

University of Mississippi

eGrove

Electronic Theses and Dissertations

Graduate School

2011

Ectomycorrhizal Fungal Succession in a Native Monterey Pine Forest and Its Potential Influence Upon Forest Population Dynamics

Kristopher Jordan Hennig

Follow this and additional works at: <https://egrove.olemiss.edu/etd>



Part of the [Ecology and Evolutionary Biology Commons](#)

Recommended Citation

Hennig, Kristopher Jordan, "Ectomycorrhizal Fungal Succession in a Native Monterey Pine Forest and Its Potential Influence Upon Forest Population Dynamics" (2011). *Electronic Theses and Dissertations*. 132. <https://egrove.olemiss.edu/etd/132>

This Dissertation is brought to you for free and open access by the Graduate School at eGrove. It has been accepted for inclusion in Electronic Theses and Dissertations by an authorized administrator of eGrove. For more information, please contact egrove@olemiss.edu.

ECTOMYCORRHIZAL FUNGAL SUCCESSION IN A NATIVE MONTEREY PINE
FOREST AND ITS POTENTIAL INFLUENCE UPON FOREST POPULATION DYNAMICS

A Thesis

Presented in partial fulfillment of requirements

for the Master of Science Degree

in the Department of Biology

The University of Mississippi

by

KRISTOPHER J. HENNIG

July 2011

ABSTRACT

Ectomycorrhizal fungi are plant symbionts influential to the growth and survival of many plant species found within forest ecosystems throughout the globe. The succession of ectomycorrhizal communities during forest maturation has been observed in a number of systems; however, though it is generally assumed that this process is important for forest dynamics, we do not fully understand the causes of succession and whether it strongly influences metrics of plant population dynamics such as population growth rates or size class structure.

I studied a secondary successional chronosequence to test the hypothesis that ectomycorrhizal fungal succession occurs within a native Monterey pine forest and parallels the trajectory observed in the well-studied bishop pine system. I also characterized the abiotic environmental factors strongly associated with successional changes in community composition. The hypothesis was partially confirmed by the increase in abundance of *Russula spp.* with increasing tree size and by the lack of association to environmental factors with the genus *Tomentella*. Furthermore, factors influential in describing the distribution of genera differed between species within these genera.

Additionally, I utilized recent findings that mature forest ectomycorrhizal fungal networks, or common mycorrhizal networks (CMN's), reduce seedling mortality and pine demographic data in the same forest type to model the potential influence of these mature forest ectomycorrhizal fungal communities on pine population growth rates and stable size class structure. I hypothesized that removal of beneficial CMN effects from forest seedlings would significantly decrease pine population growth rates. Comparisons between the projected

population growth rates from the observed demographic data and the adjusted model removing CMN benefits from seedlings indicated little difference between population growth rates or stable size class structure, thereby rejecting my hypothesis. A model scenario in which CMNs increase survivorship throughout the lifetime of this species, however, suggests that if CMNs influence the trees' entire life cycle, they are extremely important for the persistence of these forest populations. Altogether, these results suggest that ectomycorrhizal succession is occurring in this native Monterey pine forest, but its influence on plant population dynamics may depend on the extent to which CMNs influence forest trees.

DEDICATION

This work is dedicated to my parents, Ron and Joyce Hennig, who have encouraged me every step of my way and demonstrated through their actions many of the things I believe to be inherent to a good life.

LIST OF ABBREVIATIONS AND SYMBOLS

95% CI	95% confidence interval
ACE	Abundance-based coverage estimator
AICc	Akaike's Information Criterion corrected
Ca	Calcium
CEC	Cation exchange capacity
CMN	Common mycorrhizal Network
CV	Coefficient of variation
DBH	Diameter at breast height (1.4 meters)
DBLIM	Distance-based linear modeling
DBRDA	Distance-based redundancy analysis
DE	Distribution error
EM	Ectomycorrhizal fungi
G	Growth
ICE	Incidence-based coverage estimator
ITS	Internal transcribed spacer region between small and large subunit ribosomal genes
INSDB	International Nucleotide Sequence Database
KNRMR	Kenneth Norris Rancho Marino Reserve
K	Potassium
LSU	Large subunit of the ribosomal gene
Mg	Magnesium

MRT	Calculated maximum residence time spent in a size class
MSC	Mixed size class
Model A	Model removing CMN-related survival benefits attributed to only 1st (-14.4%) and 2nd year (-26.6%) age classes of size class 1, and from the 2nd year age class in size class 2 (-26.6%)
Model B	Model removing CMN-related survival benefits attributed to size class 1 (-14.4%) and size class 2 (-26.6%)
Model X	Model removing CMN-related survival benefits of -14.4% from size class 1 and -26.6% from size classes 2 through 16, but not the final size class, 17
Model Y	Model removing CMN-related survival benefits of -14.4% from size class 1 and -26.6% from all remaining size classes
M	Mortality
Na	Sodium
OP	Oak-pine
OTU	Operational taxonomic unit
P	Phosphorus
RVI	Relative variable importance
R	Contribution of recruits in the following year per individual of each size class
SSU	Small subunit of the ribosomal gene
SD	Standard deviation
SE	Standard error
St	Stasis
S	Sulfur

UAF	University of Alaska, Fairbanks
UNITE	User-Friendly Nordic ITS Ectomycorrhizal database
W_i	Akaike weight
Zn	Zinc
λ	Finite growth rate, lambda

ACKNOWLEDGEMENTS

I would like to acknowledge and thank Dr. Jason Hoeksema for all the support and wisdom he has imparted throughout this process. This experience would not have been nearly as enjoyable, nor intellectually profitable without your help. In short, I believe Jason has given me insights into being a scientist that go beyond the science. I would also like to thank my committee Drs. Tamar Goulet and Steve Brewer for all the advice, assistance, and time they have afforded me. Additionally, I would like to acknowledge the help of Don Canestro for his tireless devotion towards “facilitating science” and the insight he afforded me at my field station, the Kenneth Norris Rancho Marino Reserve. Finally, I would like to thank the “Fungus Federation of Santa Cruz” and Sigma-Xi for their generous financial support.

Many, many thanks go out to Kristin Haider, Blake Ramsby, and Bridget Piculell for their selfless field assistance, without which data collection would have been near impossible. To the aforementioned members of the Hoeksema lab and those not yet mentioned including Tiffany Bensen, Anjel Craig, T.C. Unigwe, Eugene Lukienko, Justin Murphy, Becky Brasher, and Nicole Hergott for any and all advice and comments on grants, prospecti, and presentations, thank you for all your help.

TABLE OF CONTENTS

ABSTRACT.....	ii
DEDICATION.....	iv
LIST OF ABBREVIATIONS AND SYMBOLS.....	v
ACKNOWLEDGEMENTS.....	vii
LIST OF TABLES.....	x
LIST OF FIGURES.....	xi
CHAPTER 1: ECTOMYCORRHIZAL FUNGAL COMMUNITIES, TREE AGE, AND ASSOCIATED ABIOTIC FACTORS ALONG A SECONDARY SUCCESSIONAL GRADIENT IN A NATIVE MONTEREY PINE FOREST.....	1
CHAPTER 2: THE DEMOGRAPHY OF A NATIVE MONTEREY PINE (<i>PINUS RADIATA</i>) FOREST AND THE POTENTIAL CONSEQUENCES OF ECTOMYCORRHIZAL SUCCESSION ON FOREST GROWTH.....	53
BIBLIOGRAPHY.....	88
VITA.....	97

LIST OF TABLES

Table 1-1: Relative variable importance of sampled predictor variables in relation to taxon datasets inclusive to either all ectomycorrhizal species, species occurring in >5% of samples, all genera, or genera occurring in >5% of samples.....	33
Table 1-2: Multiple partial correlations of variables of high relative variable importance (>0.3) to the first two distance-based redundancy analysis coordinate axes for datasets including all ectomycorrhizal species or genera.....	34
Table 1-3: Abundance and incidence of species identified on roots of Monterey pine in Cambria, California at the Kenneth Norris Rancho Marino Reserve in 2010 and 2011.....	35
Table 1-4: Shannon and Simpson diversity indices of plots and site.....	36
Table 1-5: Observed and estimated site richness of plots.....	36
Table 1-6: Correlational matrix of sampled environmental variables.....	39
Table 2-1: Demographic parameter estimates for matrix model size classes.....	81
Table 2-2: Estimated finite growth rate, λ , for all 18 projected models including upper and lower 95% confidence intervals.....	82
Table 2-3: Size class distribution of pines in 2009 demographic census and proportional distribution of pines in estimated stable size class structure.....	83
Table 2-4: Elasticity analysis of observed demographic data.....	86

LIST OF FIGURES

Figure 1-1: Diagram of northern and southern plots in Cambria, California in which soil, light, tree root samples, and tree size data were collected.....	32
Figure 1-2: Mau Tau species accumulation curve including 95% confidence intervals	38
Figure 1-3: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal genus-level compositional changes and the distribution of the genus <i>Russula</i>	40
Figure 1-4: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal genus-level compositional changes and the distribution of the genus <i>Wilcoxina</i>	41
Figure 1-5: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal genus-level compositional changes and the distribution of the genus <i>Rhizopogon</i>	42
Figure 1-6: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal genus-level compositional changes and the distribution of the genus <i>Tricharina</i>	43
Figure 1-7: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal genus-level compositional changes and the distribution of the genus <i>Laccaria</i>	44
Figure 1-8: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal genus-level compositional changes and the distribution of the genus <i>Tomentella</i>	45
Figure 1-9: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal species-level compositional changes and the distribution of <i>Cenococcum geophilum</i>	46

Figure 1-10: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal genus-level compositional changes and the distribution of the genus <i>Cenococcum</i>	47
Figure 1-11: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal species-level compositional changes and the distribution of <i>Russula integriformis</i>	48
Figure 1-12: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal species-level compositional changes and the distribution of <i>Russula aff sanguinea</i>	49
Figure 1-13: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal species-level compositional changes and the distribution of <i>Russula californiensis</i>	50
Figure 1-14: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal species-level compositional changes and the distribution of <i>Tomentella subililacina</i>	51
Figure 1-15: Distance-based redundancy analysis incorporating variables of high relative variable importance for ectomycorrhizal fungal species-level compositional changes and the distribution of <i>Tomentella1</i>	52
Figure 2-1: Diagram of demographic plots in which data used for modeling were collected.....	78
Figure 2-2: The average probability of a demographic event occurring for each size class (stasis, growth, mortality).....	79
Figure 2.3: Average relative contribution of recruits in following year per individual within each size class.....	80
Figure 2-4: Projected stable size class structure for the observed demographic data model, model Y, and model X.....	84

Figure 2-5: Distribution of 2009 demographic census and observed data projections of stable size class structure.....	85
Figure 2-6: Projected population growth rates of the 18 projection matrices including 95% confidence intervals.....	87

I: ECTOMYCORRHIZAL FUNGAL COMMUNITIES, TREE AGE, AND ASSOCIATED ABIOTIC FACTORS ALONG A SECONDARY SUCCESSIONAL GRADIENT IN A NATIVE MONTEREY PINE FOREST

INTRODUCTION

Mycorrhizal fungi are ubiquitous in terrestrial ecosystems throughout the world, occurring in diverse habitats from grassland and savannas to boreal, temperate, and tropical forests. The potential importance of mycorrhizal fungi to plant populations is underscored through their observed influence upon plant growth (Karst et al. 2008, Hoeksema et al. 2010), pathogen resistance (Sylvia 1983a, Borowicz 2001), drought tolerance (Allen and Boosalis 1983, Parke et al. 1983, Boyd et al. 1986), and increased nutrient acquisition (Bowen 1973, van der Heijden et al. 2006). Importantly, plants may vary tremendously in their response to different mycorrhizal fungi (Allen and Boosalis 1983, Parke et al. 1983, van der Heijden et al. 1998, Sikes and Klironomos 2009). Moreover, functional variability of different mycorrhizal species contributes to variation in ecosystem processes such as nutrient cycling (Read and Perez-Moreno 2003, Treseder 2005, Koide et al. 2007). Consequently, spatial and temporal variation in mycorrhizal community composition may drastically influence plant population dynamics, plant community structure, and ecosystem function.

The arbuscular mycorrhizal fungi, a group composed of relatively few species, associate with a vast number of herbaceous and some woody plant species generally occurring in mineral

soils with relatively low litter accumulation. In contrast, ectomycorrhizal (EM) fungal communities are often species rich (Horton and Bruns 2001), associating with woody plant communities with greater amounts of litter and, as such, are the predominant mycorrhizal fungal type within many temperate forest ecosystems (Read 1991). Although these generalities usually hold true, they belie the dynamic fungal suites and the complex biotic, environmental, and temporal relationships that structure these communities. Just as many plant communities undergo successional processes, changing in composition over time, observations of temporal transitions have been made in communities of mycorrhizal fungi. This temporal variation, or succession, of mycorrhizal fungi has been repeatedly recognized in EM forests (Fleming 1983, Fleming et al. 1985, Fox 1986, Visser 1995)

Mason et al. (1983) made the first observation of EM fungal succession while studying sporocarp occurrence over a ten-year period following the planting of a mixed stand of birches. Since then, ectomycorrhizal succession has been investigated in a number of systems; during primary succession following volcanic events (Nara et al. 2003), glacial retreat (Helm et al. 1996, Jumpponen 2003), and within sand dune plant communities (Ashkannejhad and Horton 2005); as well as during secondary succession following fire disturbance (Visser 1995, Twieg et al. 2007) and forest clear-cutting (Twieg et al. 2007). Distinct fungal communities observed on seedlings of burned and nearby unburned habitats provide additional support of EM fungal succession (Taylor and Bruns 1999).

Together, these studies and others suggest ectomycorrhizal fungi have very different life history strategies. The earliest parlance spoke of early- and late-stage fungi, i.e. species of fungi that occurred at distinct stages in forest development. However, observations that species can be found at multiple stages of forest development (Keizer and Arnolds 1994, Kranabetter et al.

2005) or in different successional seres within different forests muddies the use of temporal classifications for these fungal communities. Newton (1992) argues to inform classifications of these fungi through an understanding of their ecological functional traits that mediate EM fungal succession. Advances in our knowledge of the putative functional traits of EM fungi have been successful in some respects (Agerer 2001, Treseder 2005, Koide et al. 2007), though questions still abound as they pertain to EM succession and general EM ecology. Despite this informational gap, and the caveats about temporal classifications, there are some general characteristics that tend to distinguish pioneer and later-stage EM fungal species.

The “early-stage” or pioneer EM fungi colonize tree seedlings through spore dispersal or other resistant propagules (e.g. sclerotia) that can withstand harsh disturbance events. Spore dispersal leads to rapid relative increases of early-stage spores in the soil of disturbed sites (Deacon and Fleming 1992, Jumpponen 2003, Smith and Read 2008). Additionally, resistant propagules may lay dormant in soils, survive disturbance, and associate with root-tips of ruderal successional plants which in turn have responded to disturbance (Baar et al. 1999, Taylor and Bruns 1999). These spore banks, consisting of dispersed and disturbance resistant spores, may then be readily available for forest stand expansion into nearby habitat. This dispersal strategy poises particular EM fungi to colonize and proliferate as priority effects may play a major role in the competitive interactions that determine which mycorrhizal species establish (Kennedy et al. 2003, Kennedy et al. 2009). As the forest develops and plant root systems converge, later-stage mycorrhizal species are thought to colonize mainly through mycelial growth, connecting neighboring plant roots together in “common mycorrhizal networks” (CMNs) (Simard et al. 2002, Simard and Durall 2004).

Despite increasing discussions in the ecological literature, there is little consensus as to the primary factors that structure mycorrhizal communities during succession. Assuredly, forest plant composition influences EM fungal composition as the degree of host specificity varies amongst EM fungi (Molina 1992, Hoeksema 1995), yet even in studies of monodominant tree species, evidence of EM fungal succession has been observed (Keizer and Arnolds 1994, Visser 1995, Taylor and Bruns 1999). Various suggestions have been made to explain this observed succession. Late-stage fungi may drive their own dominance over early-stage EM fungi in the forest through colonizing root tips more competitively, having higher stress tolerance, or better resource exchange rates with plant symbionts. There is also evidence that EM fungi fall upon a continuum in their ability to access nutrients in the humus layers that may accumulate at the soil surface (Read and Perez-Moreno 2003). It is possible that forest development and ultimately the accumulation of leaf litter drive mycorrhizal succession by selecting for EM fungi that can better access increasingly recalcitrant nutrients that build up in these layers (Agerer 2001). This example shows how EM fungal succession and plant succession may directly or indirectly affect each other. Perhaps the most obvious environmental factors to change during forest maturation include decreasing levels of light due to canopy closure and increasing levels of organic matter due to deposition of forest organic matter. It is not yet clear, however, how ectomycorrhizal communities are influenced or relate to even the most apparent forest age associated environmental changes, let alone other less obvious edaphic changes.

Ideally, successional studies would follow the trajectory of a community over multiple years (Johnson and Miyanishi 2008); however, this approach is extremely time intensive, particularly when attempting to capture the full breadth of forest development. A common approach towards circumventing this problem is to study a chronosequence, whereby sites of

similar soil type and climatic and geological history are chosen so that each site differs only in the age of forest development, substituting space for time. While a number of studies have utilized a chronosequence approach to studying EM community successional dynamics through the use of multiple stands at varying ages of development, few studies have attempted to elucidate EM fungal secondary succession by sampling along a continuous ecosystem successional gradient within a given site (Yamashita et al. 2008). Investigating a successional gradient as opposed to multiple stands increases the likelihood that the composition and abundance of fungal propagules and geological history along plots will be more similar. In addition, sampling along such a gradient has the added benefit of more tightly linking changes in abiotic factors to EM fungal community composition.

In this study, I examined the belowground EM fungal community on native *Pinus radiata* trees along a gradient ranging from mature mono-dominant *Pinus radiata* forest to an area dominated by grassland vegetation into which the *Pinus radiata* forest is actively expanding since cessation of grazing. The objectives of this study were to: 1) characterize the relationship between EM fungal community composition and relative stand age and correlated abiotic factors, and 2) determine if any particular EM fungal species or genera exhibited strong relationships towards these correlated factors.

I hypothesized that EM fungal succession within this *Pinus radiata* forest would resemble that observed in studies conducted within bishop pine (*Pinus muricata*) forests. These forests may share similar successional taxa for a number of reasons. First, these forests are separated by less than 450 km and are ecologically restricted to similar Mediterranean climate types with equitable levels of precipitation, the majority of which falls between the months of December and March. Secondly, these species are sister taxa, each exhibiting a degree of

serotiny, suggesting a similar natural fire regime that may select for similar EM fungal communities (Gardes and Bruns 1996, Rogers 2002). Lastly, observations of similar aboveground EM fungal communities (K. Hennig and J. Hoeksema, personal observations) suggest that the belowground component may follow a similar trajectory (Gardes and Bruns 1996). Specifically, I hypothesized that taxa of *Rhizopogon*, *Tuber*, and *Wilcoxina* would be abundant in the grassland (early-stage) community and members of *Amanita* and *Russula* would dominate the mature (late-stage) forest community, with purported multi-stage species including *Cenococcum geophilum* and species of *Tomentella* appearing in abundance along the successional gradient.

The series of successional studies conducted on bishop pine are among the best examples of EM fungal succession so far (Gardes and Bruns 1996, Horton and Bruns 1998, Taylor and Bruns 1999, Bruns et al. 2005). Still, we understand little about what factors are driving this process, even in these well-studied forests. Moreover, we know very little about the ectomycorrhizal communities within native Monterey pine forests. Only five native Monterey pine forests remain, acting as habitat for a unique complement of flora and fauna (Rogers 2002), yet Monterey pine is one of the most planted pine species in the world and known to be an aggressive invader in non-native habitats. Broadening our knowledge of EM fungal successional dynamics in these Monterey pine forests may allow land managers and conservationists to further generalize common characteristics of successional studies or utilize specific knowledge of succession in these forests to develop more effective conservation and management practices for at-risk forest populations.

MATERIALS AND METHODS

Study site

The study site was located in Cambria, California on the Kenneth Norris Rancho Marino Reserve (KNRMR) (35° 32' 25.21" N, 121° 05' 29.57" W). Cambria, California is host to one of the five remaining populations of native Monterey pine (*Pinus radiata*) with two residing to the north and south of Cambria along the coast of California and the remaining two upon islands off the coast of Baja California, Mexico. This region receives ~460 mm of annual precipitation (Canestro Pers. Comm.), intermediate amongst the three coastal stands of Monterey pine forest in California (Rogers 2002). The forest soil consists mainly of deep sandy loam (Carpenter and Storie 1933) which is low in organic matter (8-15 cm) and easily erodes. Mature Monterey pines in Cambria average 30-37 m tall, may root as deep as soil or clay permit (~ 1.7 m in deep soil), and have extensive lateral spread (9-12 m). Monterey pine is the dominant tree species with only one other hardwood associate: Coast Live Oak (*Quercus agrifolia*), which is also ectomycorrhizal.

In moist areas, common understory plants are bracken (*Pteridium sp.*), California blackberry (*Rubus ursinus*), and poison oak (*Toxicodendron diversilobum*) while drier sites include coast sagebrush (*Artemisia californica*), coyotebrush (*Baccharis pilularis*), and bush monkeyflower (*Mimulus aurantiacus*). At the forest edge, the understory consists primarily of grasses including California oat grass (*Danthonia californica*), Blue wild rye (*Elymus glaucus*), Purple needle grass (*Nasella pulchra*), Kikuyu grass (*Pennisetum clandestinum*), and Harding grass (*Phalaris aquatica*) (McDonald and Laacke 1990, Canestro Pers. Comm.).\

Data Collection

Biotic and abiotic variables

I selected two plots (36 m x 65 m), each spanning the transition from grassland to forest interior representing a secondary vegetational chronosequence (Figure 1-1). Within each of these plots, I collected a total of 140 soil cores (15 cm deep x 1.5 cm diameter) in January 2010, with one taken every 5 meters along the Y-axis (perpendicular to the vegetational gradients) and every 4 meters along the X-axis (parallel to the vegetational gradient) to characterize a variety of soil and light characteristics. Along each of the 14 lateral 36 m ('X-axis') transects in each plot, I pooled together the 10 soil samples, and passed them through a 2 mm sieve into a basin. Soil subsamples of each pooled set were sent to the ANR Analytical Laboratory at the University of California, Davis and the Mississippi State University Cooperative Extension Soil Analysis Laboratory for determination of soil organic matter percent, organic carbon percent and extractable amounts of Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg), Zinc (Zn), Sulfur (S), and Sodium (Na) and cation exchange capacity (CEC). Soil texture was also characterized for each pooled sample (Lamotte soil texture test kit). Additional abiotic data collected in January 2010 for each of the separate 140 core locations included canopy cover, litter depth, and soil gravimetric water content. Subsequent sampling was conducted in January 2011 to collect additional soil moisture data, as well as pH and soil texture, for each of the separate 140 core locations. pH has been found to be relatively invariant in conifer stands of similar types over periods ranging from eight to nearly forty years (Kranabetter et al. 2005, Marcos et al. 2007). Therefore, I expect that pH data would not have changed appreciably during the time between the collection of abiotic factors in 2010 and those collected in 2011.

Canopy cover was assessed with canopy photography using a Nikon Coolpix 990 digital camera fit with a fisheye lens. Image analysis of these photographs was conducted with Gap Light Analyzer software (Frazer et al. 1999) allowing estimates of percent canopy openness and total light penetration. Litter depth was determined, using a measuring stick, as height from the mineral soil to the top of the litter layer. Soil water content in 2010 was recorded as the average of three individual water content measurements using a 5 cm theta probe, each taken 5 cm from the soil core site and equidistant from each other. In 2011, soil water content was determined gravimetrically for core samples of the upper 5 cm of soil.

Finally, during collection of the ectomycorrhizal samples (described below), tree height (for trees less than 200 cm tall) and diameter at breast height (DBH = 140 cm) were recorded. These tree height and DBH data were used to determine relative size classes ranging from 1 – 17. These relative size classes were constructed using demographic data collected in the same Cambria forest and were based upon the mortality, reproductive effort, and growth rates of Monterey pine trees ranging in size from 3 to 140 cm in height and 93 cm DBH (Hennig et al. Unpublished).

Ectomycorrhizal collection

From the 4 x 5 meter area surrounding each soil core point in each plot, I collected a root sample from all Monterey pines less than 200 cm in height and from up to one individual greater than 200 cm in height. For pines less than 20 cm in height, the entire plant was collected. Root samples from pines greater than 20 but less than 200 cm in height were collected by digging down at the base of the tree, identifying up to three primary roots, and excising a 15 cm portion starting 5 cm down the length of that root. Roots from trees greater than 200 cm in height were

sampled by coring within the drip line of the tree, but at least 1 m away from any other individuals. All collected soil cores, excised root segments, and seedlings were placed in individual zip lock bags with field soil and were shipped to the University of Mississippi where they were stored in a refrigerator at 4⁰ C until processing. Roots were hand washed over a 2 mm sieve and the root-tip region encompassed by the fungal hartig net of the EM fungi was collected. Up to ten mycorrhizal root tips were randomly removed with the use of a dissecting microscope and placed in individual tubes for immediate DNA extraction.

Perhaps due to a relatively long and dry summer in 2010 (Canestro Pers. Comm.), pine seedling abundances in the grassland were lower than expected. Consequently, eight additional seedlings (8 – 33cm tall) were collected in the grassland in January of 2011. Although statistical analysis of these samples with the samples collected in 2010 along the entire vegetational gradient would not be appropriate, I collected these additional samples to inform our understanding of how EM fungal communities during the earliest successional stages of forest encroachment into the grassland may differ from those during later stages of forest development.

Molecular identification of ECM fungi

DNA was extracted from root tip samples using the Sigma Extract-N-Amp extraction kit (Sigma-Aldrich, St. Louis, MO), as follows: to each root tip, 10 µl of the Sigma Extraction Buffer was added, and then the sample was heated to 65⁰C for 10 minutes and 95⁰C for 10 minutes, after which 30 µl of the Sigma Neutralization Solution was added. The extraction yielded DNA at a concentration of 387.1 (SD±344.2) ng/µl that was diluted to 40% by adding 60 µl of PCR-grade water. Polymerase Chain Reaction (PCR) was then performed using the fungal-specific forward and reverse primers, NSI1 and NLB4 (Martin and Rygiewicz 2005). These

primers amplify the internal transcribed spacer regions (ITS1 and ITS2) between the small subunit (SSU) and large subunit (LSU) ribosomal genes. This DNA region rapidly evolves in the fungal genome and thus allows for effective species level determination (Gardes and Bruns 1993). Each 8 μ l PCR reaction contained 0.4 μ l (10 μ M concentration) of each primer, 4 μ l of 2X REDExtract-N-Amp PCR Reaction Mix, 2.7 μ l of sterile PCR-grade water, and 0.5 μ l of a 154.8 (SD \pm 137.7) ng/ μ l DNA sample. Therefore, the final concentration of DNA for each PCR reaction was 9.7 (SD \pm 8.6) ng/ μ l per sample. Thermocycling for PCR included the following provisions: the initial denaturation for 3 minutes at 94 $^{\circ}$ C, followed by 30 cycles of denaturation for 45 seconds at 94 $^{\circ}$ C, annealing for 45 seconds at 58 $^{\circ}$ C and extension for 72 seconds per cycle at 72 $^{\circ}$ C, followed by a final extension time of 10 minutes at 72 $^{\circ}$ C. The PCR reactions were checked for amplification on a 1 % agarose gel with SYBR $^{\circ}$ Safe DNA gel stain (Molecular Probes, Eugene, OR, USA). Excess primer and unincorporated nucleotides were removed enzymatically using ExoSAP-IT (USB Corporation Cleveland, OH, USA) with the following procedure: 0.25 μ l ExoSAP-IT and 4.75 μ l sterile PCR-grade water were added to 5 μ l of the PCR product and incubated at 37 $^{\circ}$ C for 45 minutes, then 80 $^{\circ}$ C for 20 minutes, and finally 4 $^{\circ}$ C for at least 5 minutes.

Sequencing was performed using the forward primer ITS1F (Gardes and Bruns 1993) and the Big Dye Terminator Sequencing Kit (v3.1, Invitrogen Corp.) Each Big Dye reaction contained 0.4 μ l Big Dye Reaction Premix, 1.8 μ l Big Dye 5 X sequencing buffer, 0.5 μ l of the forward primer in 10 μ M concentration, and 6.3 μ l of PCR-grade water. Nine μ l of Big Dye mastermix was distributed into each well with 1 μ l of the cleaned PCR product. Thermocycling conditions were as follows: 96 $^{\circ}$ C for 1 minute followed by 35 cycles of 95 $^{\circ}$ C for 30 seconds, 50 $^{\circ}$ C for 20 seconds, and 60 $^{\circ}$ C for 4 minutes (Applied Biosystems recommended). Reactions

were dried and shipped overnight to the DNA Lab at Arizona State University, in Tempe, Arizona, where the Big Dye reactions were purified and read on a capillary genetic analyzer.

The fungal sequences were edited manually in Geneious software (Biomatter Ltd.), correcting ambiguous bases associated with dye blobs and elsewhere when possible. All sequences with >3% ambiguous bases and < 200 base pairs long were deleted. These remaining sequences were subjected to operational taxonomic unit (OTU) assembly (at 97% similarity) using Cap3 software (Huang and Madan 1999) on the University of Alaska, Fairbanks (UAF) informatics server, as described previously (Taylor et al. 2007) using default settings except the following: maximum overhang percent length = 60, match score factor = 6, overlap percent identity cut-off = 96, clipping range = 6. Grouping homologous sequences that are >97% similar as a specific OTU is a conservative approach employed by previous studies (Izzo et al. 2005, O'brien et al. 2005, Smith et al. 2007) that assumes a .2%-1.2% error rate produced by PCR and unidirectional sequencing, as well as ~1.5% divergence of the ITS region that may occur within some species at small spatial scales (Horton 2002). The consensus fungal sequences from each OTU were submitted using BLAST (nucleotide) searches on the International Nucleotide Sequence Database (INSD), User-Friendly Nordic ITS Ectomycorrhizal (UNITE) database, and the curated fungal ITS database on the UAF Informatics Portal to obtain best matches for taxonomic affiliation of OTUs. The ultimate decision on the best match to a sequence was based on both similarity and length of the match. Sequences >97% similar in composition to database sequences from named, cultured fungi were considered the same OTU (hereafter, 'species').

Sequences with matches <97%, but >94% similarity to a database sequence with an assigned species epithet, or matching a sequence identified only to genus, were placed into the

respective genus and given a number (e.g. *Russula* 1). Similarly, those matches in the database <94%, but greater than 90% were assigned to the appropriate taxonomic family. Any matches <90% similar to database sequences were excluded from the analyses.

If sequence matches among the three sequence repositories showed equal affinity or similarity to multiple genera within a family, priority was given to the vouchered specimens residing on the UNITE or curated fungal ITS databases. Any species known to be strictly non-mycorrhizal was eliminated from the data set.

Statistical Analysis

Richness and diversity

Measures of richness and diversity were calculated using EstimateS Version 8.2. All identified species were used to estimate diversity for each plot separately, and were also pooled to estimate species richness of the whole field site in a separate analysis. For these diversity estimates, I included both “Singlets” (species recovered only once) and “Contigs” (species recovered more than once). Mean site-level Shannon and Simpson diversity indices were determined by pooling together samples from both plots. Comparisons between the plots for these same parameters were calculated by first rescaling to the greatest shared number of individuals (n=124). Additional non-parametric richness based estimators were also calculated, including Chao 1 and 2, Jackknife 1 and 2, abundance-based coverage estimator (ACE), and incidence-based coverage estimator (ICE). Chao’s estimated coefficient of variation (CV) for abundance and incidence distributions equaled 0.993 and 0.692, respectively. Chao and Lee (1992) suggest that when the CV is > 0.5, the higher estimate between Chao 1 and ACE for abundance-based data and between Chao 2 and ICE for incidence-based data is the more precise

estimate of species richness. For that reason, ACE and ICE are reported. Uncertainty (SD) of these non-parametric estimates was calculated through 50 randomizations of sample order.

To gauge whether sampling intensity was sufficient to accurately describe the ectomycorrhizal community, species accumulation curves with 95% confidence intervals were also generated. The Mau Tau function was used to generate species accumulation curves which describe the number of ectomycorrhizal species retrieved as a function of the number of ectomycorrhizal root tips successfully identified.

Distance-Based Linear Modeling

Permanova was conducted prior to pooling taxonomic data from the two plots together for distance-based linear modeling to ensure that the fungal taxonomic composition of each plot did not differ significantly (Pseudo-F 1,71 = 0.96, $p=0.411$). A distance-based linear modeling (DBLIM) approach was employed to determine the factors most influential on EM fungal community composition using four variations of the community data:

1. All observed species (including all singlets and contigs)
2. Species that occurred in >5% of the samples
3. All observed genera (i.e. all species belonging to a particular genus were grouped together)
4. Genera that occurred in >5% of the samples

For DBLIM, I modeled a matrix composed of Bray-Curtis similarity scores of EM fungal community composition among samples as a function of multiple predictor variables. In total I collected data on 19 predictor variables, which were either pooled across each lateral 36 m row

of soil cores (soil organic matter percent, organic carbon percent, and extractable amounts of P, K, Ca, Mg, Zn, S, Na, soil texture (clay, silt, and sand percent) and CEC) or unpooled (pH, soil gravimetric water content in 2010 and 2011, percent canopy openness, litter depth, relative tree size (1-17)). Percent organic carbon and silt percent were left out from analyses because either they were highly correlated (>99%) with soil organic matter percent or the remaining soil texture estimates (Clay and sand %) were perfect correlates, respectively.

Two different approaches to analysis were used on each of the 4 datasets listed above. First, DBLIM analyses were conducted on all predictor variables after averaging the collected unpooled predictor variables across each lateral 36 m X-axis row of soil cores, creating additional “pooled” predictor variables to run in the analysis with the other pooled variables. This form of the analysis had much less power to detect variations in community abundances due to many fewer data points (280 down to 28). Therefore, I conducted additional analyses with all pooled and unpooled variables; however, this time I made the simplifying assumption that each value for the pooled variables along each X-axis row of soil cores was the same for each of the 10 points along that row. While this approach creates pseudoreplication, I felt it was important to understand the general relationship between many of these environmental variables (P, K, Ca, Mg, Zn, S, Na, CEC, and soil texture (Sand % and Clay %)) that are infrequently used in determining relationships to community variation. The results of these two sets of analyses were very similar; therefore, I present only the results from the latter analyses utilizing both pooled and unpooled variables.

For each analysis of the four variant community datasets, I utilized all 17 predictor variables in generating 500 models with different combinations of those 17 predictor variables. Those models were ranked according to an information-criterion, AIC_c (Akaike’s Information

Criterion corrected), which corrects for small sample sizes but converges upon the AIC for large size samples. For each model, an Akaike weight (w_i) was calculated, the weight being reflective of the likelihood that the model is the best among those being considered. The relative variable importance (RVI) of each predictor variable across all models was determined through the summation of the Akaike weights of all models in which that predictor variable occurred. Authors have suggested various minimum values for ascribing importance to RVI values that have ranged from 0.3 - 0.5 (out of a possible value up to one). For purposes of distance-based linear model analysis, I have chosen to use the less conservative of these values, 0.3, to ensure that important patterns are not missed when comparing environmental variation to community composition.

Distance-Based Redundancy Analysis

Distance-based redundancy analysis (DBRDA) was used to depict relationships between variables with high RVI (from the DMLIM analyses described above) and distributions of the species and genera found within the plots. For this purpose, I plotted dataset 1 including all observed species against the set of variables with high RVI (>0.3) determined from DBLIM analyses for both dataset 1 (all observed species) and dataset 2 (Species that occurred in $>5\%$ of the samples) (Tables 1-1, Table 1-2). Similarly, I plotted dataset 3 (All observed genera) against the set of variables with high RVI (>0.3) determined from DBLIM analyses for both dataset 3 (All observed genera) and dataset 4 (Genera that occurred in $>5\%$ of the samples) (Tables 1-1 and 1-2).

RESULTS

Fungal taxa present in Cambria, CA at the Kenneth S. Norris Rancho Marino Reserve

A total of 367 individual root tips were identified from the sampling of 2010 (267) and 2011 (82) representing 25 ectomycorrhizal contigs and 18 singlets (43 distinct ectomycorrhizal species) as well as 6 fungal species (18 individuals) suspected of saprophytic or pathogenic origin that were removed from the data analysis. Approximately 39% of the species collected in 2010 matched accessions in one of the queried databases to a specific species, 77% to genus, 83% to family, 96% to class and order, and 100% to phylum. *Thelephoraceae* (41.1%), *Russulaceae* (34.1%), *Pyronemataceae* (17.6%), and *Rhizopogonaceae* (7.1%) were the most abundant fungal families. Genera of relatively high abundance included *Tomentella* (6 OTUs, 32.6%), *Russula* (7 OTUs, 21.3%), *Wilcoxina* (1 OTU, 13.1%), and *Rhizopogon* (3 OTUs, 5.2%). *Tomentella subulilacina* was the most dominant species in the plots (25.9%), followed by a number of *Russula* species including *R. integriformis* (12.9%), *R. californiensis* (9.4%), *R. aff sanguinea* (5.9%), *Wilcoxina*1 (10.6%), *Laccaria amethystina* (8.2%), *Cenococcum geophilum* (5.9%), and *Tomentella*1 (5.9%) (Table 1-3).

Of the eight seedlings collected in 2011, ectomycorrhizal root tips were successfully amplified from seven. While all three members of the genus *Rhizopogon* recovered in 2010 were also recovered in 2011, the relative abundance of *Rhizopogon* in the 2010 samples was low (7.1%). In contrast, *Rhizopogon* spp. were found in 100% of the root samples collected in 2011. *R. occidentalis* appeared in 57.1%, *R. roseolus* 42.8%, and *R. brunsi* 14.3%. In addition, I recovered *Wilcoxina*1 and *L. amethystina*, each from one core (Table 1-3).

Richness and Diversity

Shannon and Simpson diversity estimates suggest a diverse and relatively heterogeneous community of ectomycorrhizal fungi (Table 1-4). A high number of distinct “singlet” fungal species were found in both plots, 12 and 16 from the northern and southern plots, respectively, both sites sharing a total of 15 species. The higher number of “singlet” species found within the southern plot is despite the fact that more root tips were identified in the northern plot.

Richness based estimations were largely in concordance with one another (ACE, ICE, Jackknife 1, Jackknife 2). The Mau Tau function as well as 95% CI’s failed to reach an asymptote, so the Mau Tau function and all richness based estimators suggest that additional sampling would have revealed additional species (Table 1-5, Figure 1-2).

Distance-Based Linear Modeling

The AICc derived RVI scores for the four sets of community data showed similarities in the degree of importance for the tested predictor variables, but also clear differences. All variables with high RVI scores (> 0.3) except relative tree age and extractable Zn levels were significantly correlated with distance along the established successional gradient (Table 1-1, Table 1-6).

Tree stage (or relative age) was a relatively strong predictor of community variation in all datasets, but its relative importance differed between the data sets parsed by species or genera. Community composition at the genus level (Table 1-1, Datasets 3 and 4) was most strongly influenced by tree stage followed by extractable Ca and Zn levels. In contrast, the community composition of both species datasets exhibited similarly strong relationships to tree stage as they did to either soil moisture and Ca in 2010 (Table 1-1, Dataset 1) or soil pH (Table 1-1, Table, Dataset 2)

Generally, besides tree stage, the variables affecting species vs. genus level composition were different with a few exceptions. The variation in community composition of highly abundant species (Table 1-1, Dataset 2) was strongly influenced by soil pH, but this same variable was one of the least influential on composition of fungal genera (Table 1-1, Dataset 3 and 4). Soil moisture in 2010 and extractable P content of the soil were moderately important to the composition of highly abundant species (Table 1-1, Dataset 2). Similarly, soil Ca and soil moisture in 2010 was moderately associated with dataset 1 including all species.

In addition to tree stage, genus composition was highly associated with soil Ca and Zn (Table 1-1, Dataset 3 and 4). Moreover, composition of abundant fungal genera (Table 1-1, Dataset 4) was highly associated with percent organic matter content and CEC of the soil, and the percent of canopy openness showed a moderate effect on the composition of fungi at the genus level (Table 1-1, Dataset 3).

Distance-based redundancy analysis

Of the several genera that were most abundant, a few exhibited strong relationships to environmental factors highly correlated to distance along the environmental gradient, while others were uncorrelated with environmental variation. *Russula* appeared to be strongly associated with trees at later stages of development (Figure 1-3), being rarely found on trees smaller than 10 cm in diameter (2 of 29 instances). In contrast, *Wilcoxina*, *Rhizopogon*, and *Tricharina* showed clearer association to trees of earlier developmental stages, higher levels of Ca, and higher percent canopy openness (Figure 1-4, Figure 1-5, Figure 1-6). *Laccaria* appeared moderately associated with higher percentages of organic matter and with trees in later stages of development (Figure 1-7). *Tomentella*, in contrast, showed little association with any of the

sampled environmental variables (Figure 1-8) while *Cenococcum* and *C. geophilum* demonstrated an association to trees of moderate size or increasing Zn levels (Figure 1-9, Figure 1-10).

Variables that were influential towards distribution of genera were not always the same between species within a genus. While *Russula* as a whole was largely influenced by relative tree age, *R. integriformis* was the only species of that genus that maintained this strong association (Figure 1-11). *R. aff sanguinea* (Figure 1-12) appeared more strongly associated with increasing soil pH and Ca, while *R. californiensis* (Figure 1-13) showed little association with any plotted environmental variables. Likewise, *Tomentella subulacina* (Figure 1-14) demonstrated slight association with trees of increasing relative age while *Tomentella1* (Figure 1-15) appeared to show a slight relationship for increasing levels of extractable P and soil moisture content.

DISCUSSION

Diversity and Richness

Shannon and Simpson diversity indices reflect a highly diverse ectomycorrhizal community (Table 1-4). This is a commonly observed phenomenon within many other monodominant native forests (Horton and Bruns 2001). Interestingly, these indices are greatly above those found in a study of ectomycorrhizal communities found within a Monterey pine plantation in New Zealand. Despite extensive sampling at multiple ages of plantation development, Walbert et al. (2010) observed a grand total of 19 species colonizing the pines and calculated a Shannon and Simpson diversity index of 2.2 and 0.8; respectively. The higher diversity of the ectomycorrhizal community found within my site may reflect a more abundant

community in this native site or, alternatively, the high genetic diversity in native Monterey pine stands (Rogers 2004) could reflect the capability of genotypically different trees of the same species to harbor different ectomycorrhizal complements (Sthultz et al. 2009).

The high values of ACE, ICE, and Jackknife 1 & 2 richness estimates in comparison to Mau Tau estimates likely reflect the high weight these estimates place upon the occurrence of singlet and doublet (that appear twice) species that occur in site dataset (Table 1-5, Figure 1-2). Regardless, all richness estimates indicate that continued sampling would have recovered additional species from within my plots. This result parallels the findings of multiple studies (e.g. Horton and Bruns 1998, Horton et al. 1999, Stendell et al. 1999, Twieg et al. 2007) including even those investigating fine scale ectomycorrhizal distributions with hyper-intensive sampling efforts (Tedersoo et al. 2003).

Compositional relationships to bishop pine systems and successional status

The community of EM fungi within the plots at KNRMR was moderately consistent with successional studies conducted in forests composed of the sister species bishop pine. Many of the highly abundant genera ($\geq 5\%$ of samples) found within my plots including *Tomentella*, *Russula*, *Rhizopogon*, and *Wilcoxina* are dominant components of the bishop pine system found within either the resistant sporebank or mature forest communities (Table 1-3)(Gardes and Bruns 1996, Taylor and Bruns 1999). Specifically, my hypothesis that species of *Tuber*, *Rhizopogon*, and *Wilcoxina* would appear in greater abundance further from the forest, *Amanita* and *Russula* further into the forest, and *C. geophilum* and *Tomentella* along the entire successional gradient was only partially confirmed. *Rhizopogon*, *Wilcoxina*, and *Tricharina* were correlated with decreasing tree size and increasing light and calcium levels suggesting these taxa were often

found in the grassland. *Russula spp.* were highly correlated with increasing tree size and organic matter and decreasing light levels indicating a higher presence with increasing relative forest age. Finally, while *Tomentella* showed little association to any factors along the chronosequence, *C. geophilum* showed a moderate correlation to increasing Zn levels and mid-sized trees.

Putative early-stage taxa

While species of *Rhizopogon* (3), *Wilcoxina* (1), and *Tuber* (1) were observed upon roots of pines within my plots, none were abundant enough within samples from 2010 to justify any declarations of successional status; therefore, it would be difficult to say, on these data alone, that the “early-stage” EM fungal assemblages of this Monterey pine forest resemble the bioassay and post-fire studies of bishop pine early-stage spore bank communities (Horton et al. 1998, Taylor and Bruns 1999, Grogan et al. 2000). It is important to note, however, that in DBRDA analysis, *Rhizopogon*, *Wilcoxina*, and *Tricharina* were associated with environmental characteristics that were negatively and significantly correlated with distance along the environmental chronosequence, indicating that these taxa recovered in my plots were found in the grassland, and very often were found on trees of lesser size, suggestive of an early-stage classification (Figure 1-4, Figure 1-5, Figure 1-6). Four additional independent lines of evidence also suggest that some of these genera comprise at least part of the pioneering early-stage species of this forest and are consistent with genus level assemblages of the bishop pine forests. First, the 2011 sampling, which focused solely on grassland seedlings (<33 cm tall) greater than 10 meters away from the drip line of any other trees, recovered a complement of species almost entirely consisting of the purported “early-stage” genera (*Rhizopogon* (3 sp.) and *Wilcoxina* (1 sp.)). Second, in accordance with this 2011 sampling, a recent bioassay study of the spore bank

community in Cambria was largely dominated by an unknown *Wilcoxina* and *Rhizopogon* species (Hoeksema et al. Unpublished data). Third, DBRDA plotting of *Wilcoxina*, the only relatively abundant (>5% of samples) “early-stage” genus to be recovered, demonstrates a moderate tendency to associate with trees of lesser relative age and decreasing levels of light (Figure 1-4). This association suggests that *Wilcoxina* was found more frequently in the grassland, on the roots of younger trees, where P levels were generally higher. Finally, despite abundant inoculum of *Rhizopogon* spp. and other “early-stage” species within the soils of these coastal pine populations (Taylor and Bruns 1999, Kernaghan et al. 2003) as evidenced by the need to employ dilution methods to retrieve rare or infrequent species in the bioassays, these species are infrequently found, if at all, in surveys of mature forest fungal communities (Gardes and Bruns 1996, Horton and Bruns 1998, Taylor and Bruns 1999). If these species are not ruderal, early-stage species in these forests as well, it is surprising that the greater sampling of trees roots further into the forest interior in this study did not recover more of these species.

If the genera *Rhizopogon* and *Wilcoxina* are, as suggested by these lines of evidence, members of the early-stage sporebank assemblages of Cambria, California, it appears that the diversity and richness of this early-stage community may be relatively low compared to that of most, but not all, bishop pine systems (Kjoller and Bruns 2003). Indeed, they may be more similar in these metrics to forests of another pine, bristlecone pine (*P. longaeva*), in which soil bioassays have recovered a very species poor, but similar complement of spore bank fungi (Bidartondo et al. 2001). Additionally, sampling of root tips in 2011, though replete with colonization of *Rhizopogon* and *Wilcoxina*, failed to recover any additional species within these genera from those found in 2010, suggestive of low diversity. Alternatively, this result may indicate higher abundances or stronger competitive abilities of these taxa versus other possible

“early-stage” taxa in this environment. This lack of spore-bank diversity is in agreement with bioassay work from this Cambria site which suggests that among the five native Monterey pine populations, Cambria soils ranked lowest in both estimated richness and Shannon diversity of EM sporebank species (ACE = 9.78 (SD=4.37), Shannon diversity = 0.62 (SD = 0.10); Hoeksema et al. Unpublished data). In conjunction, Kjoller and Bruns (2003) *Rhizopogon*-centric study of soils from four bishop pine populations suggested that diversity of early-stage species within this region of California may be generally low. This idea is supported by additional evidence that members of the *Pinus* clade are typically capable of associating with a relatively full complement of *Rhizopogon* species, suggesting that it is not the specificity of *P. radiata* to particular *Rhizopogon* species that limits their observed diversity in this survey (Rusca et al. 2006).

Putative late – stage taxa

Although I observed no ectomycorrhizal root-tip colonization of *Amanita* species within my plots, db-RDA plots of *Laccaria* indicates a relatively strong association with trees of increasing age and increasing percent organic matter suggesting a placement in the late-stage community of this forest (Figure 1-7). This finding parallels those by Gherbi et al. (1999), observing high genetic diversity and presence of *Laccaria amethystina* within a 150-year-old beech forest.

Additionally, there was strong support for my hypothesis regarding the late-successional status of *Russula* species. *Russula*, second in abundance only to *Tomentella*, appeared to be a diverse genus at this site, with three relatively dominant taxa (*R. integriformis*, *R. californiensis*, and *R. aff sanguinea*) and four additional species appearing only once or twice within my plots.

Russula was strongly associated with decreasing levels of canopy openness suggesting that this genus increased in abundance with increasing distance into the forest (Figure 1-3). Additionally, and importantly, relative tree age appears to greatly contribute to the observed compositional changes to *Russula* (Figure 1-3). These observations concur with studies of bishop pine (Gardes and Bruns 1996, Horton and Bruns 1998, Taylor and Bruns 1999) and other mono-dominant or mixed-forests (Horton and Bruns 1998, Kernaghan et al. 2003, Douglas et al. 2005, Aponte et al. 2010) where *Russula* is among the most diverse and dominant taxa at later stages of forest development.

While these EM fungal successional studies and others have observed similar increases in abundance or diversity of *Russula* with increasing stand age, many of these studies have been conducted upon even-aged forests stands or “tree islands” (Visser 1995, Smith et al. 2002, Kranabetter et al. 2005, Twieg 2006, Twieg et al. 2007), preventing the separation of the effects of soil properties and stand age from the effect of tree age in which root-tips were collected. EM community composition may be partially influenced by tree age or size, as opposed to stand age, through direct biotic influences such as tree age-specific variation in mycorrhizal available carbon amounts (Deacon and Fleming 1992) or indirectly by influencing the soil composition of mycorrhizal helper bacteria (Garbaye and Duponnois 1992, Garbaye 1994) through age-related differences in quality or quantity of root exudates (Grayston and Jones 1996). Alternatively, root densities have been implicated in influencing EM fungal composition (Peay et al. 2010). Even-aged stands may have more uniform root densities as opposed to the mixed-age stands of my site, so this strong association of tree age to genus composition (Table 1-1, Table 1-2) may partly be a reflection of the age structure of my plots.

Putative multi-stage taxa

Tomentella (6 spp.) was frequently found within my plots (32.6% of samples), and notably, *T. sublilacina* was the most abundant (22.8% of all identified root-tips) and frequently found (25.9% of samples) EM fungal species. This is a common theme of bishop pine studies in which *T. sublilacina* often represents a large component of the EM fungal community (Gardes and Bruns 1996, Horton et al. 1998, Taylor and Bruns 1999). The notable dominance of *Tomentella* in the plots and its apparent lack of relationship to factors commonly associated with forest age such as tree age, percent canopy openness, or percent organic matter is generally supportive of *Tomentella*'s multi-stage status (Figure 1-8).

The placement of *C. geophilum* in this system as a multi-stage species is preliminary. DBRDA plots including variables of high RVI (>0.3) for species (Figure 1-9) or genera (Figure 1-10) showed similar patterns; specifically, *C. geophilum* showed a mild relationship to trees of intermediate developmental stages and intermediate levels of percent organic matter. This pattern is more suggestive of an EM fungal taxon associated with moderate levels of stand development than one occurring at multiple stages of forest development; however, the relatively low numbers of *C. geophilum* recovered in this study beg more work in this system to bolster these observations.

Genus and species responses to environmental factors

Relative tree age proved to be the factor with the strongest association to changes in EM fungal community composition along the gradient. The high degree of importance placed upon tree age relative to sampled edaphic factors (Table 1-1, Table 1-2) and clear association of some EM fungal taxa (e.g., *Russula*) with trees of greater relative age was strongly indicative of a

successional progression within this site. This result parallels the findings of research conducted in multiple systems (Last et al. 1987, Keizer and Arnolds 1994, Visser 1995, Taylor and Bruns 1999, Twieg et al. 2007, Yamashita et al. 2008). These compositional changes may be explained by increasing photosynthetic rates and associated availability of carbon that is often concurrent with increasing tree age (Deacon and Fleming 1992). In consequence, ingress by species with greater carbon demands, but better competitive abilities may occur. Similarly, increasing inoculum availability of vegetatively dispersed species (those species often categorized as “late-stage”) in proximity to older trees could also help explain the observed shifts in abundance of some taxa within my site (Deacon and Fleming 1992).

In addition to tree age, various edaphic properties were also implicated as important aspects to understanding community composition (Table 1-1, Table 1-2). Many of the variables with high RVI were significantly correlated with increasing distance along the environmental gradient supporting the argument that these ectomycorrhizal fungal communities are undergoing successional changes (Table 1-1, Table 1-6). Interestingly, different variables were important for explaining community composition of highly abundant species versus genera. For instance, pH was more important than relative tree age for highly abundant species, while it was less important compared to most other factors for composition of genera.

Russula is an exemplar of this phenomenon. As a genus, it demonstrated strong associations with trees of greater relative age (Figure 1-3); however, DBRDA analysis of *Russula* spp. suggested that different factors explain distributions of different *Russula* species during succession. Specifically, while *R. integriformis* was consistently found in association with trees of greater relative age (Figure 1-11), *R. aff sanguinea* appeared to correspond strongly to increasing pH and soil Ca (Figure 1-12) and conversely, *R. californiensis* showed little

association to any sampled environmental factors (Figure 1-13). While it is difficult to confidently extrapolate from the observed patterns of only three species within this genus, these differences may indicate that partitioning of the soil environment is acting to facilitate coexistence among these closely related species. This observation could help explain the sequential addition and increasing abundance of *Russula* species with increasing stand-age observed in multiple studies (Visser 1995, Kranabetter et al. 2005, Twieg et al. 2007).

In similar fashion, *Tomentella*, the most abundant genus within the plots, displayed very little relationship to environmental variation within the plots (Figure 1-8). However, after DBRDA plotting of both *Tomentella* species, *Tomentella*1 shows greater association to trees of intermediate developmental stages and decreasing levels of extractable P (Figure 1-15) while *T. sublilacina* displays a modest association to trees of greater relative age and increasing percent organic matter (Figure 1-14). This result, however, does not conflict with *T. sublilacina*'s purported multi-stage status. Lilleskov and Bruns (2003) observed that *T. sublilacina* demonstrated an ability to germinate from spores which are not common in most mature forests. In conjunction, its lack of receptivity to nutrient additions and yet steady increase in abundance during the course of their study indicated that a competitive strategy including initial spore dormancy, early spore germination subsequent to disturbance, followed by vegetative colonization may ultimately lead to community dominance.

Soil chemistry and its novel role in community composition

It is worth noting the surprising finding that two soil factors most associated with compositional changes of genera, abundant or otherwise, and highly abundant species included

levels of extractable Ca and Zn (Table 1-1, Table 1-2). There is a general dearth of information on how these elements may act to affect ectomycorrhizal communities in natural environments.

DBRDA of three highly abundant genera (>5% of samples) within my plots including *Tricharina*, *Rhizopogon*, and *Wilcoxina* demonstrated clear associations of these taxa for higher Ca conditions, and it may have been these associations that caused Ca to be highly important in explaining community structure. Aponte et al. (2010) observed strong influences of leaf litter and soil concentrations of Ca, as well as soil pH, upon EM fungal community composition, which they believed to be indirectly mediated by host-specific leaf-litter compositions. Ultimately, the authors suggested that changes in soil acidity, modified by Ca influences upon exchangeable base saturation, were responsible for EM fungal species distribution. In my analyses, pH displayed low importance in both models of genus composition, suggesting that a similar association between Ca and pH is unlikely in this instance. In contrast, highly abundant species were strongly associated with pH and CEC, a closely related measure to exchangeable base saturation, and so an indirect influence of Ca on pH may be a viable explanation for explaining some species distributions. Alternatively, soil Ca in this study was significantly correlated ($r=-0.64$, $p < 0.0001$) with distance along the environmental gradient, and so may have been strongly correlated with some unmeasured forest age associated variable influential to EM fungal community composition (Table 1-1, Table 1-6).

Finally, it is known that Zn and other metals are essential for proper fungal growth and metabolism, though all metals will elicit negative fungal responses at some threshold concentration (Fomina et al. 2005b). The majority of *in vitro* studies examining the effect of Zn or Zn-containing minerals upon EM fungi have used concentrations greatly exceeding those levels found on unpolluted, natural environments (Blaudez et al. 2000, Belling-Abler

2004, Fomina et al. 2005a, Fomina et al. 2005b). One study, however, conducted at Zn levels comparable to my field site (0 – 3 ppm) demonstrated inhibition and stimulation of EM fungal growth as well as interspecific variation in EM fungal response even at these low Zn concentrations (Corguz et al. 2010).

CONCLUSION

EM fungal successional studies have been instrumental in advancing how we view the relationships between EM fungi and forest age and correlated environmental variation. The successional gradient approach taken in this study is novel in that fine scale sampling along this gradient enables us to detect genera and species changes during forest maturation, and how environmental factors associated with increasing stand age correspond to these compositional changes. This study suggests that tree age and other soil properties such as Ca, Zn, percent organic matter, and CEC are influential to the community composition of EM fungal genera. Importantly, however, these relationships may or may not be the same for specific fungi within the same genus. This result indicates that some degree of soil niche partitioning may be contributing to the coexistence of closely related species, but that different factors contribute to niche partitioning at the genus level. It will be important for researchers to be cognizant of this possibility when deciding whether to pool taxa together for purposes of analysis.

Finally, the importance of sampling a variety of environmental factors not generally thought to be explanatory of EM fungal composition is highlighted by the finding that Ca and Zn are variables strongly associated with EM fungal distributions at my site. The interactions and auto-correlations of soil factors associated with increasing stand age make it difficult to pinpoint the causal factor in this relationship. In addition, unmeasured variables correlated with these soil

properties may be partly responsible for this change. By identifying important, and perhaps unexpected relationships between soil properties and EM community composition, and species distributions, as has been observed here, we might develop subsequent experiments to unveil the causal mechanisms or functions behind these relationships.

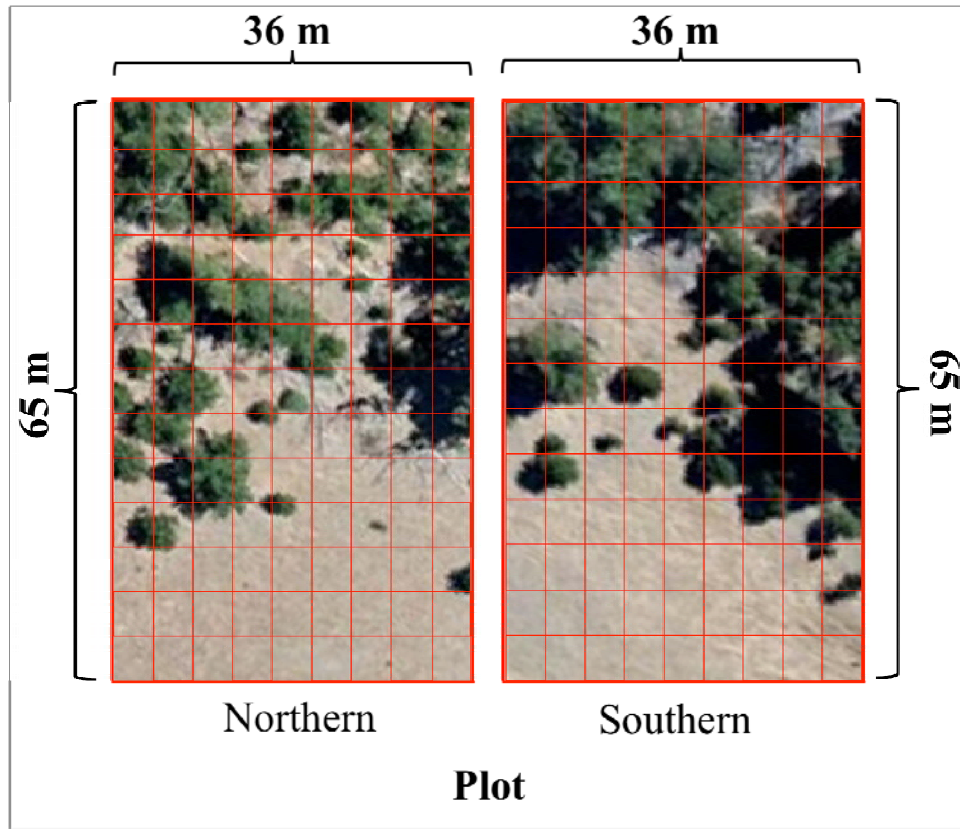


Figure 1-1: Northern and southern plots in which environmental and ectomycorrhizal data was collected. At each intersecting red line, environmental data characterizing light and soil information was collected. Additionally, in a five m² rectangle surrounding this point, ectomycorrhizal root samples were collected from one adult tree and every tree ≤ 2 meters tall. Finally, tree size data was recorded from the tree in which root samples were collected.

Table 1-1: Relative variable importance scores determined by generating the best 500 models, assigning each a weight representing the likelihood that that is the best model, and assigning the summed value to each variable of those models that it was a part of. Therefore, higher relative variable importance values represent the variables presence in more and better models. The first column consists of those seventeen predictor variables I used in the distance-based linear modeling. Additionally, the four center columns represent the four variations of community data that were analyzed separately. The farthest right column gives the correlation of each variable to distance along the environmental chronosequence and associated p-value. A positive correlation indicates a positive association with increasing distance into the forest. An asterisk (*) following a predictor variables name indicates a high relative variable importance score (≥ 0.3) in one of the four distance-based linear modeling analyses and included in distance-based redundancy analysis for either species or genus level distributions.

Predictor Variable	1. All species	2. Species (>5%)	3. All genera	4. Genera (>5%)	Correlation to distance along the environmental chronosequence (P-value)
Tree stage*	0.268	0.517	0.941	.971	0.01 (0.93)
2010 H2O*	0.323	0.272	0.194	.162	-0.52 (<0.0001)
2011 H2O	0.051	0.028	0.029	.015	0.18(0.09)
pH*	0.255	0.644	0.140	.113	-0.46(<0.0001)
Litter depth (mm)	0.084	0.022	0.214	.284	0.43(<0.0001)
Canopy Openness (%)*	0.166	0.075	0.355	.183	-0.66(<0.0001)
Organic Matter (%)*	0.169	0.355	0.268	.443	0.35(0.001)
P (ppm)*	0.186	0.325	0.198	.259	-0.42(<0.0001)
K (ppm)	0.088	0.150	0.081	.113	-0.03(0.76)
Ca (ppm)*	0.329	0.474	0.535	.728	-0.68(<0.0001)
Mg (ppm)	0.079	0.087	0.061	.099	0.01(0.91)
Zn (ppm)*	0.083	0.218	0.389	.606	0.13(0.23)
S (ppm)	0.225	0.193	0.285	.238	0.73(<0.0001)
Na (ppm)	0.187	0.111	0.055	.084	0.13(0.23)
CEC*	0.111	0.168	0.314	.454	0.28(0.01)
Sand (%)	0.052	0.012	0.129	.100	-0.39(0.0002)
Clay (%)	0.213	0.191	0.045	.070	0.49(<0.0001)

Table 1-2: Multiple partial correlations of variables of high relative variable importance as determined by distance-based linear modeling to the first two distance-based redundancy analysis coordinate axes for datasets including all sampled ectomycorrhizal species or genera. Best model variables included in distance-based redundancy analysis for all species included tree stage, 2010 H₂O, pH, percent organic matter, extractable P (ppm), and Ca (ppm). Best model variables included in distance-based redundancy analysis for all genera included tree stage, percent canopy openness, percent organic matter, Ca (ppm), Zn (ppm), and cation exchange capacity (CEC).

Predictor Variable	Variable correlation to first two db-RDA axes (All Species)		Variable correlation to first two db-RDA axes (All Genera)	
	db-RDA 1	db-RDA 2	db-RDA 1	db-RDA 2
Tree stage	-0.372	0.480	0.707	-0.265
2010 H₂O	0.434	-0.233		
2011 H₂O				
pH	0.445	0.549		
Litter depth (mm)				
Canopy Openness (%)			-0.401	0.340
Organic Matter (%)	-0.336	0.201	0.301	0.094
P (ppm)	0.468	-0.235		
K (ppm)				
Ca (ppm)	-0.336	0.564	0.190	0.736
Mg (ppm)				
Zn (ppm)			-0.429	-0.173
S (ppm)				
Na (ppm)				
CEC			-0.168	-0.483

Table 1-3: OTU-group names (species) identified on roots of Monterey pine in Cambria, California at the Kenneth Norris Rancho Marino Reserve. The four right-most columns show the total number and percent of root-tips (abundance) or samples (incidence) in which these OTU-groups were found in both 2010 and 2011.

OTU-group name	bp	Best match (Accession #)	% Similarity (bp)	# Abundance 2010 (2011)	% Abundance 2010 (2011)	# Incidence 2010 (2011)	% Incidence 2010 (2011)
<i>Ascomycota1</i>	225	<i>Ascomycota</i> sp. (DQ822805)	96.0 % (216)	1	0.37	1	1.18
<i>Ascomycota2</i>	460	Uncultured <i>Ascomycota</i> (EF619866)	97.2 % (447)	1	0.37	1	1.18
<i>Atheliaceae1</i>	554	<i>Atheliaceae</i> sp. (GU180260)	99.6 % (553)	10	3.75	2	2.35
<i>Cenococcum geophilum</i>	647	<i>Cenococcum geophilum</i> (AY394919)	99.4 % (644)	7	2.62	5	5.88
<i>Ceratobasidiaceae1</i>	423	Uncultured <i>Ceratobasidiaceae</i> (HM141009)	96.0 % (409)	1	0.37	1	1.18
<i>Ceratobasidiaceae2</i>	569	Uncultured <i>Ceratobasidiaceae</i> (HM141007)	95.6 % (546)	1	0.37	1	1.18
<i>Cortinarius aff pauperculus</i>	445	<i>Cortinarius aff pauperculus</i> (GQ159858)	98.2 % (438)	2	0.75	1	1.18
<i>Cortinarius rigens</i>	747	<i>Cortinarius rigens</i> (GQ159809)	97.2 % (731)	1	0.37	1	1.18
<i>Dothideomycetes1</i>	308	Uncultured <i>Dothideomycetes</i> (DQ273305)	97.1 % (299)	1	0.37	1	1.18
<i>Hebeloma1</i>	580	<i>Hebeloma</i> sp. KGP23 (DQ822802)	99.1 % (574)	1	0.37	1	1.18
<i>Helotiales1</i>	559	<i>Helotiales</i> sp. P224 (FN669205)	96.6 % (542)	5	1.87	3	3.53
<i>Helotiales2</i>	345	Uncultured <i>Helotiales</i> (FM995595)	97.1 % (335)	1	0.37	1	1.18
<i>Helotiales3</i>	396	Uncultured <i>Helotiales</i> (EU649083)	96.7 % (382)	1	0.37	1	1.18
<i>Helotiales4</i>	446	Uncultured <i>Helotiales</i> (FJ440902)	96.7 % (432)	1	0.37	1	1.18
<i>Helotiales5</i>	648	Uncultured <i>Helotiales</i> (DQ497974)	94.2 % (621)	1	0.37	1	1.18
<i>Helvella lacunosa</i>	609	<i>Helvella lacunosa</i> (HQ650748)	98.2 % (599)	6	2.25	4	4.71
<i>Hydnoplicata1</i>	505	<i>Hydnoplicata</i> sp. src701 (DQ974734)	96.8 % (489)	2	0.75	2	2.35
<i>Hydnoplicata2</i>	505	<i>Hydnoplicata</i> sp. src701 (DQ974734)	98.6 % (498)	1	0.37	1	1.18
<i>Inocybe geophylla</i>	608	<i>Inocybe geophylla</i> isolate (HQ604291)	99.7 % (606)	2	0.75	2	2.35
<i>Inocybe mixtilis</i>	327	<i>Inocybe mixtilis</i> (HQ604493)	97.9 % (320)	1	0.37	1	1.18
<i>Laccaria amethystina</i>	797	<i>Laccaria amethystina</i> (EF530940)	100 % (797)	12 (1)	4.49 (1.22)	7 (1)	8.24 (14.29)

Table 1-3 (Continued): OTU-group names (species) identified on roots of Monterey pine in Cambria, California at the Kenneth Norris Rancho Marino Reserve. The four right-most columns show the total number and percent of root-tips (abundance) or samples (incidence) in which these OTU-groups were found in both 2010 and 2011.

OTU-group name	bp	Best match (Accession #)	% Similarity (bp)	# Abundance 2010 (2011)	% Abundance 2010 (2011)	# Incidence 2010 (2011)	% Incidence 2010 (2011)
<i>Rhizopogon brunsii</i>	651	<i>Rhizopogon brunsii</i> (AY971832)	100 % (651)	2 (11)	0.75 (13.41)	2 (1)	2.35 (14.29)
<i>Rhizopogon occidentalis</i>	809	<i>Rhizopogon occidentalis</i> (DQ822821)	98.9 % (805)	3 (48)	1.12 (58.54)	1 (4)	1.18 (57.14)
<i>Rhizopogon roseolus</i>	580	<i>Rhizopogon roseolus</i> (GQ179949)	99.8 % (579)	9 (12)	3.37 (14.63)	3 (3)	3.53 (42.86)
<i>Russula aff sanguinea</i>	605	<i>Russula aff sanguinea</i> UC 18 (EU248591)	100 % (605)	12	4.49	5	5.88
<i>Russula californienis</i>	628	<i>Russula californiensis</i> (AY245542)	99.7 % (627)	14	5.24	8	9.41
<i>Russula integriformis</i>	680	<i>Russula integriformis</i> (AY061684)	98.2 % (678)	25	9.36	11	12.94
<i>Russula xerampelina</i>	662	<i>Russula xerampelina</i> (AY061734)	99.7 % (660)	3	1.12	2	2.35
<i>Russula1</i>	233	<i>Russula aff sanguinea</i> UC 18 (EU248591)	96.6 % (225)	1	0.37	1	1.18
<i>Russula2</i>	309	Uncultured <i>Russula</i> (EU668264)	97.1 % (300)	1	0.37	1	1.18
<i>Russula3</i>	321	Uncultured <i>Russula</i> (FJ013095)	97.2 % (313)	1	0.37	1	1.18
<i>Suillus pungens</i>	620	<i>Suillus pungens</i> (L54094)	99.4 % (618)	1	0.37	1	1.18
<i>Thelephora1</i>	630	<i>Thelephora regularis</i> (U83485)	96.2 % (606)	2	0.75	2	2.35
<i>Tomentella sublilacina</i>	804	<i>Tomentella sublilacina</i> (DQ482017)	100 % (804)	61	22.85	22	25.88
<i>Tomentella1</i>	741	<i>Tomentella</i> sp. (EF411099)	95.6 % (718)	13	4.87	5	5.88
<i>Tomentella2</i>	734	<i>Tomentella</i> sp J54 (AJ534914)	94.0 % (690)	8	3.00	2	2.35
<i>Tomentella3</i>	576	<i>Tomentella</i> sp Src822 (DQ974780)	94.5 % (545)	3	1.12	2	2.35
<i>Tomentella4</i>	437	<i>Tomentella lateritia</i> (DQ974777)	95.2 % (418)	1	0.37	1	1.18
<i>Tomentella5</i>	451	<i>Tomentella sublilacina</i> (DQ482002)	96.7 % (438)	1	0.37	1	1.18
<i>Tricharina1</i>	416	<i>Tricharina</i> sp. (EU726332)	94.7 % (397)	3	1.12	2	2.35
<i>Tricharina2</i>	648	<i>Tricharina</i> sp. (EU726332)	96.9 % (633)	7	2.62	4	4.71
<i>Tuber californicum</i>	340	<i>Tuber californicum</i> (DQ974799)	98.8 % (336)	2	0.75	2	2.35
<i>Wilcoxina1</i>	643	<i>Wilcoxina</i> sp (GU181904)	96.3 % (628)	35 (10)	13.11 (12.20)	9 (1)	10.59 (14.29)

Table 1-4 – Shannon and Simpson diversity indices. Northern and southern plot values based upon the highest number of shared individuals between both plots (n = 124). Standard deviations of Shannon and Simpson diversity are based on variation in sample order among randomizations and so all equal 0 when considering all collected samples.

Site	Shannon	Simpson
Pooled Plots	2.93	0.089
Northern Plot	2.56	0.125
Southern Plot	2.82	0.085

Table 1-5 – Estimations of site richness. 95% confidence intervals of Mau Tau represent the range of fungal richness to be expected if another sampling of similar intensity within this site was undertaken. Displayed ACE, ICE, Jackknife 2 and Jackknife 2 mean values based upon bootstrapped runs of the data. Standard deviations of ACE, ICE, and Jackknife 2 are based on variation in sample order among randomizations and so all equal 0 when considering all collected samples.

	Mau Tau (Obs)	Mau Tau lower 95% CI	Mau Tau upper 95% CI	ACE	ICE	Jackknife 1 (SD*)	Jackknife 2
Pooled Plots	43.0	36.5	49.5	68.1	67.2	61.8 (4.1)	68.8

*Calculated analytically

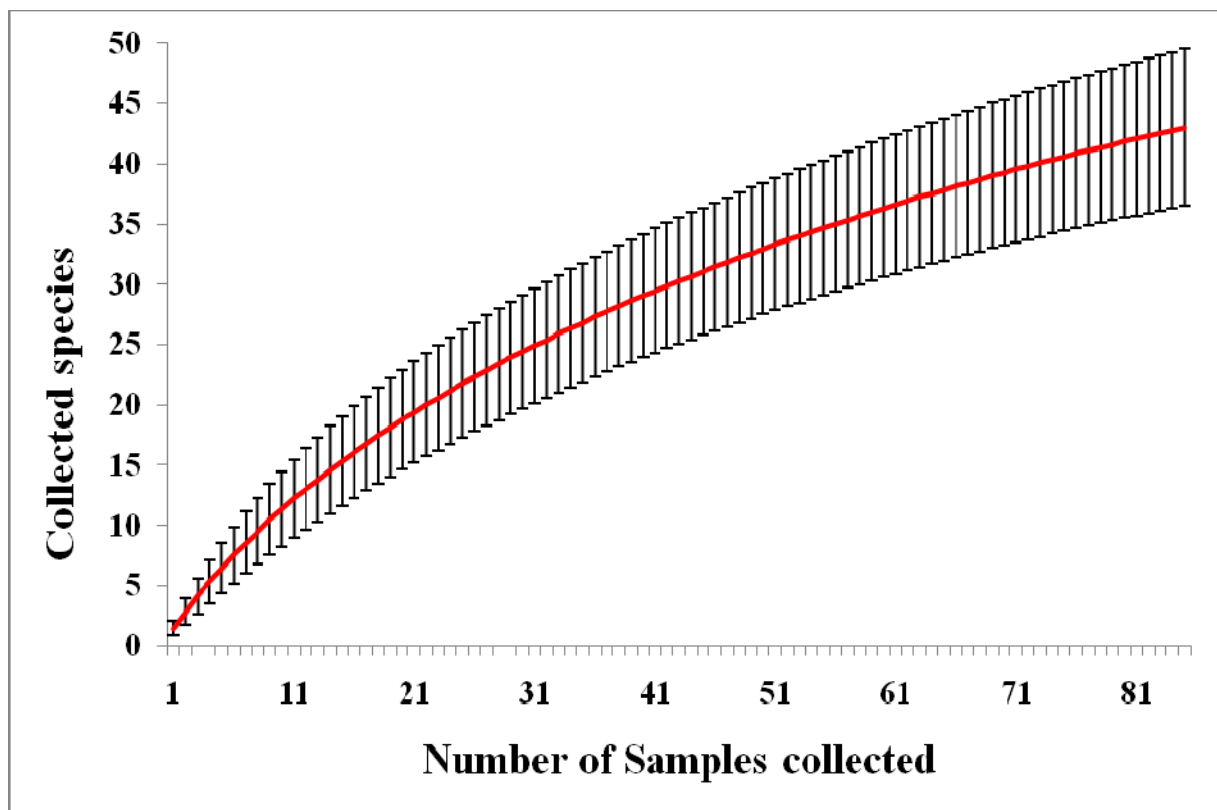


Figure 1-2 – Mau Tau species accumulation curves. Red line represents the average number of species collected with n collected root samples in site plots. Upper and lower 95% confidence (black lines) intervals estimate the number of species that could be expected to be collected with an identical sampling strategy in these plots. Average and 95% confidence intervals have been calculated by bootstrapping collected samples fifty times.

Table 1-6: Correlational matrix of collected environmental variables. (RTA = relative tree age, DIF = distance along transects spanning from grassland into the forest, 2010 H₂O = gravimetric soil moisture in 2010, 2011 H₂O = soil moisture content in 2011, pH, LD = litter depth (mm), % CO = percent canopy openness, % OM = percent organic matter, P = phosphorus, K = potassium, Ca = calcium, Mg = magnesium, Zn = zinc, S = sulfur, Na = sodium, CEC = cation exchange capacity, % Sand = percent sand content of soil, % Clay = percent clay content of soil)

	RTA	DIF	2010 H ₂ O	2011 H ₂ O	pH	LD	% CO	% OM	P	K	Ca	Mg	Zn	S	Na	CEC	% Sand
DIF	0.01																
2010 H2O	-0.21	-0.52															
2011 H2O	-0.24	0.18	-0.17														
pH	0.06	-0.46	0.42	-0.34													
LD	0.15	0.43	-0.25	0.01	-0.12												
% CO	-0.37	-0.66	0.35	0.10	0.14	-0.39											
% OM	-0.03	0.35	-0.36	0.31	-0.47	0.01	-0.06										
P	0.08	-0.42	0.32	0.06	0.10	-0.24	0.25	0.07									
K	0.25	-0.03	0.03	-0.28	0.07	-0.02	-0.25	0.04	0.49								
Ca	-0.07	-0.68	0.49	0.05	0.09	-0.32	0.45	0.13	0.46	0.05							
Mg	-0.16	0.01	0.16	0.19	-0.29	0.06	0.03	0.61	0.26	0.13	0.56						
Zn	0.00	0.13	-0.02	0.30	-0.43	-0.05	-0.02	0.61	0.62	0.34	0.38	0.64					
S	-0.11	0.73	-0.39	0.32	-0.55	0.31	-0.31	0.62	-0.18	-0.02	-0.18	0.34	0.48				
Na	-0.03	0.13	0.24	-0.14	-0.01	0.15	-0.35	0.19	0.22	0.31	0.19	0.68	0.28	0.09			
CEC	-0.09	0.28	-0.07	0.36	-0.51	0.05	-0.08	0.78	0.32	0.10	0.40	0.83	0.88	0.66	0.42		
% Sand	-0.01	-0.39	0.18	-0.05	0.28	-0.27	0.36	-0.25	-0.15	-0.43	0.12	-0.35	-0.37	-0.37	-0.50	-0.37	
% Clay	0.29	0.49	-0.32	-0.08	-0.25	0.24	-0.49	0.41	0.05	0.48	-0.24	0.32	0.36	0.26	0.35	0.35	-0.49

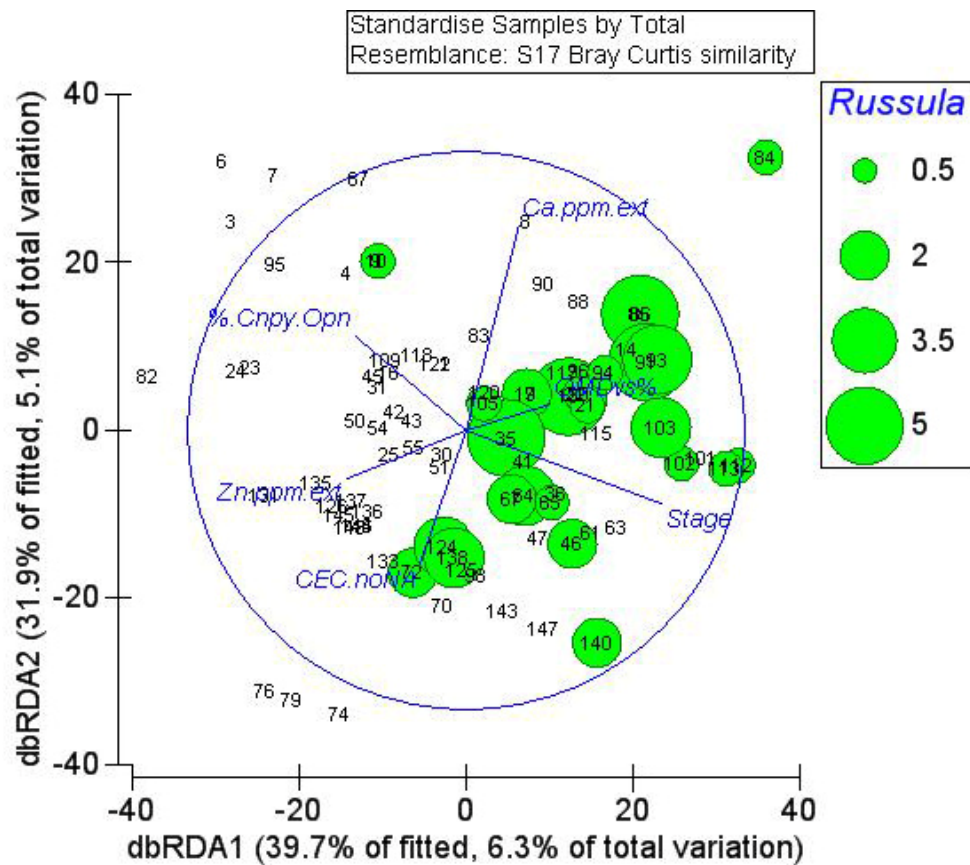


Figure 1-3 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both genus level datasets (All genera and genera occurring in $>5\%$ of samples and the distribution of *Russula* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Russula* within that sample.

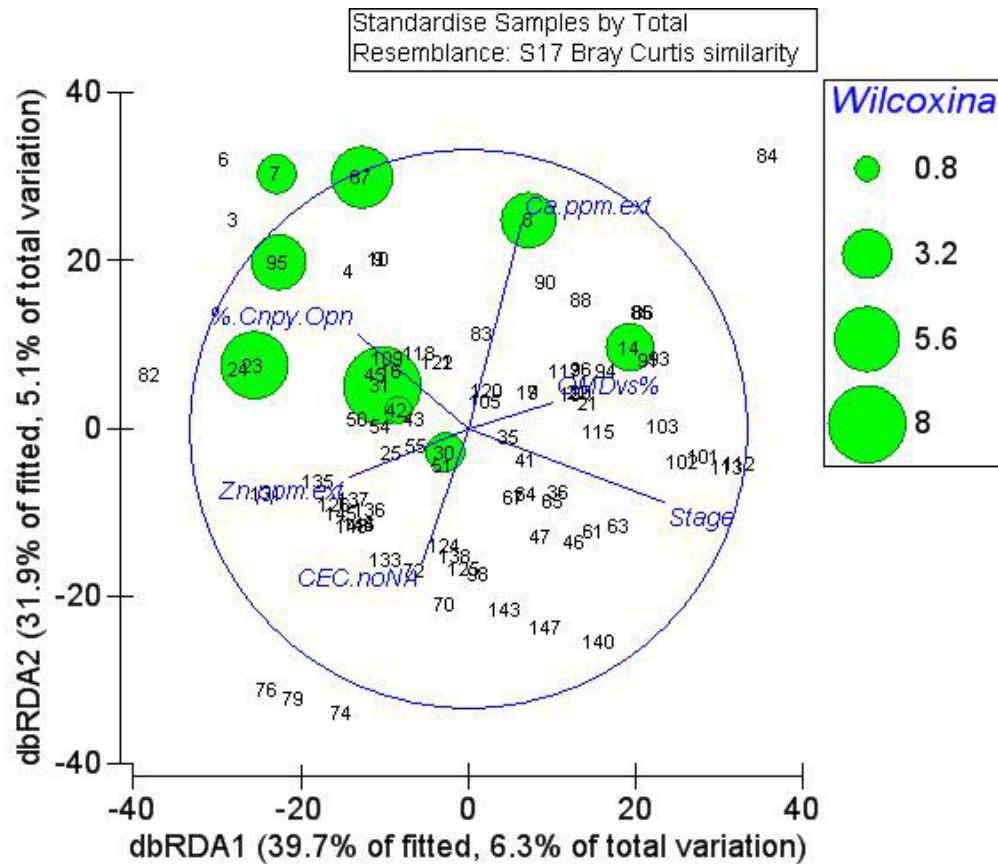


Figure 1-4 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both genus level datasets (All genera and genera occurring in $>5\%$ of samples and the distribution of *Wilcoxina* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Wilcoxina* within that sample.

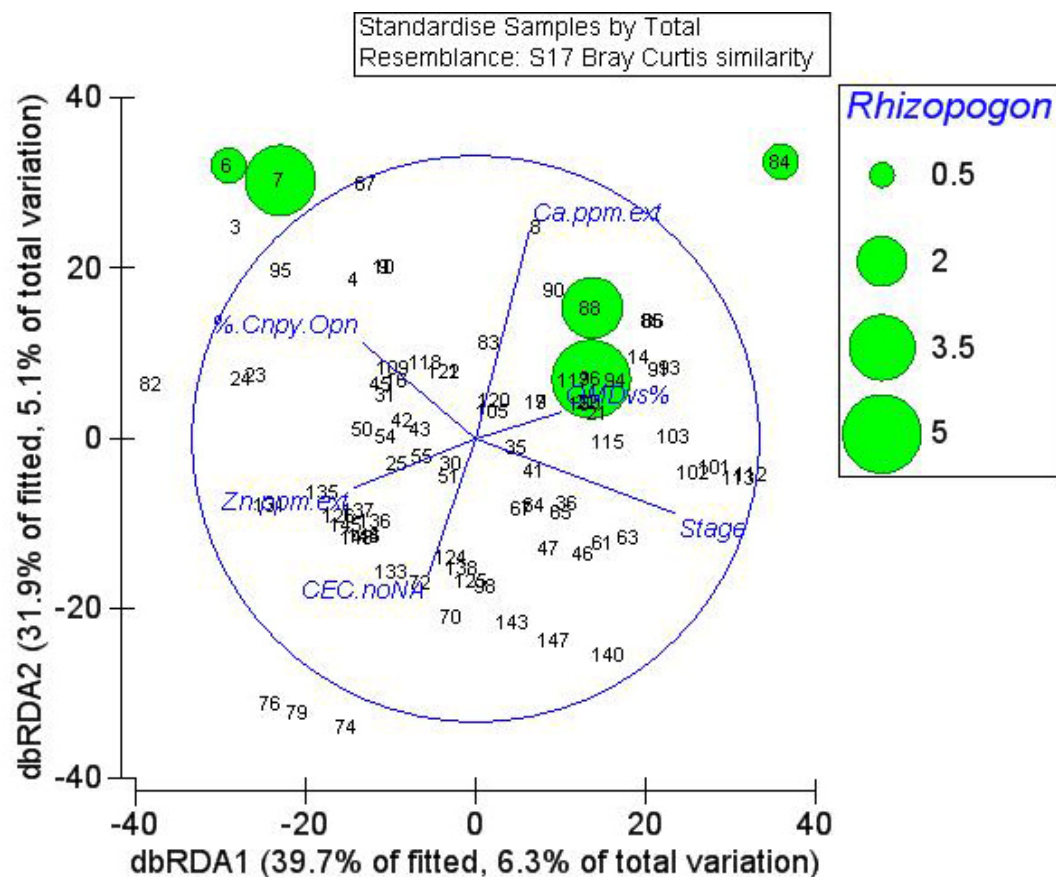


Figure 1-5 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both genus level datasets (All genera and genera occurring in $>5\%$ of samples and the distribution of *Rhizopogon* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Rhizopogon* within that sample.

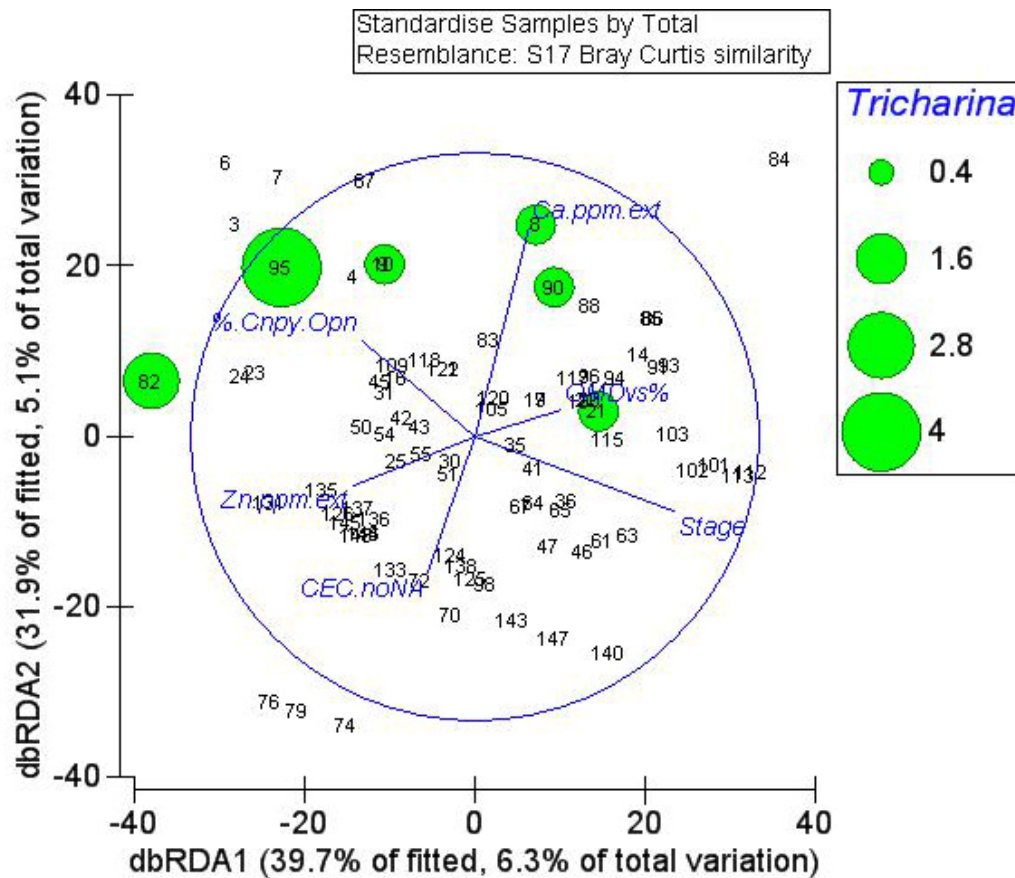


Figure 1-6 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both genus level datasets (All genera and genera occurring in $>5\%$ of samples and the distribution of *Tricharina* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Tricharina* within that sample.

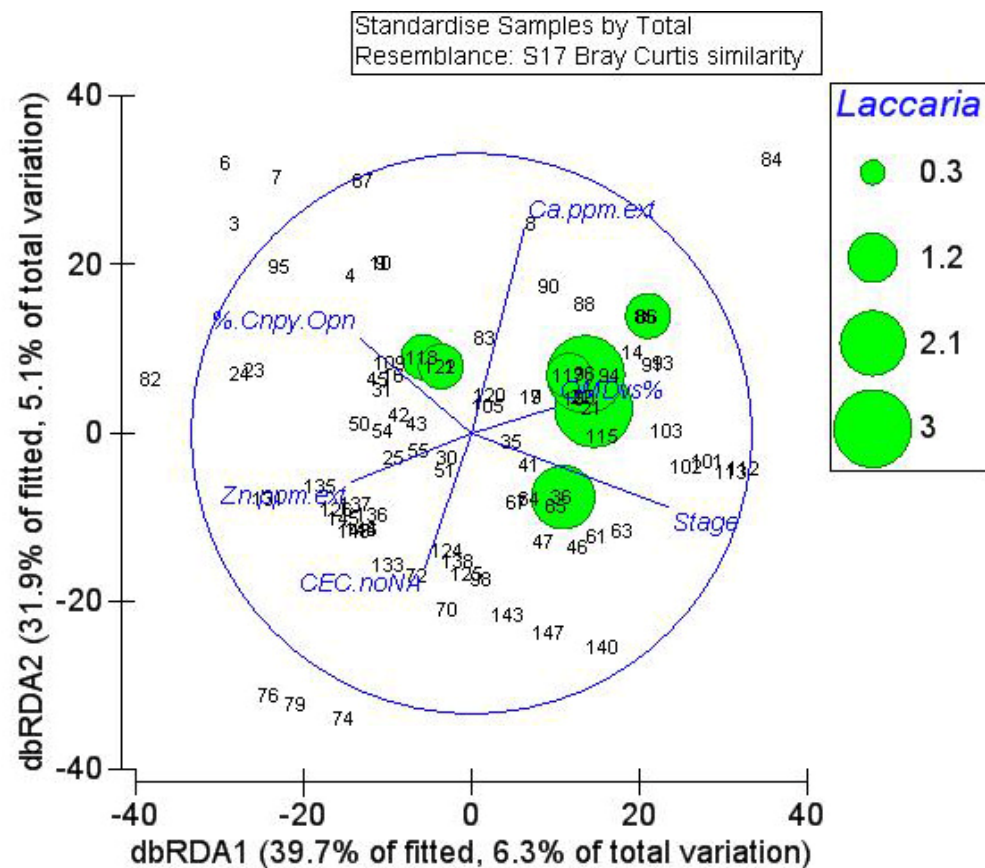


Figure 1-7 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both genus level datasets (All genera and genera occurring in $>5\%$ of samples and the distribution of *Laccaria* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Laccaria* within that sample.

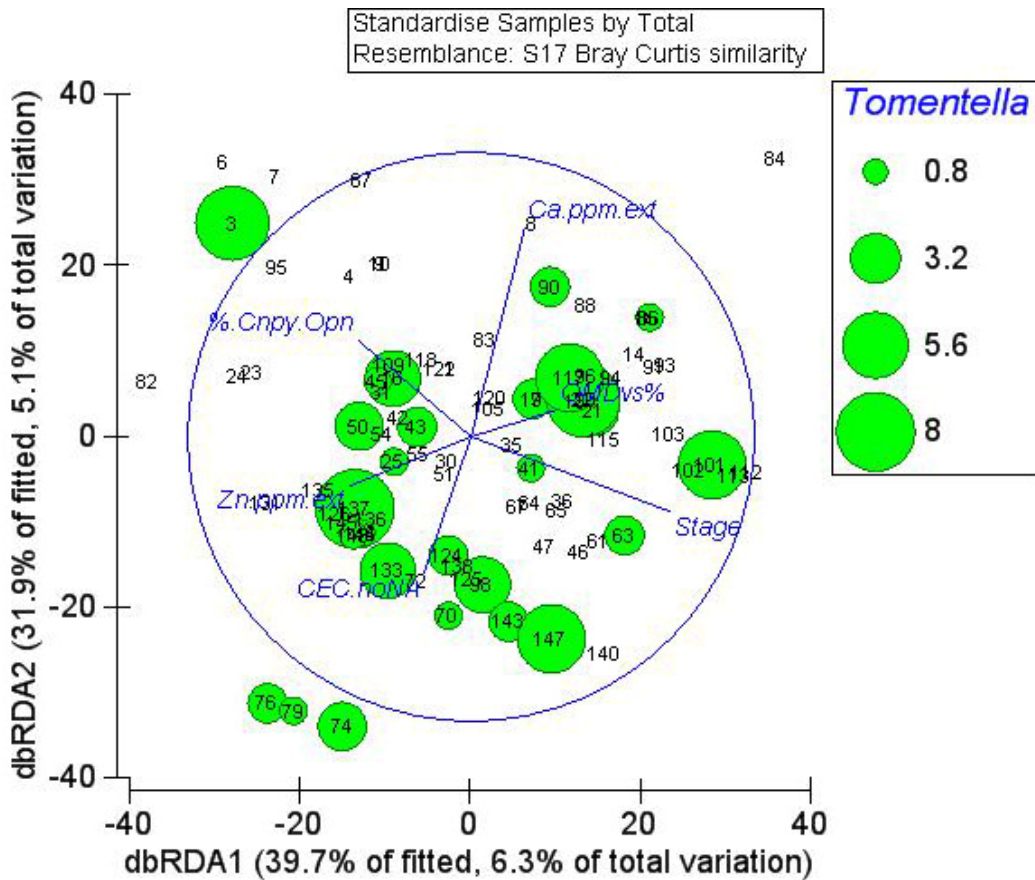


Figure 1-8 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both genus level datasets (All genera and genera occurring in $>5\%$ of samples and the distribution of *Tomentella* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Tomentella* within that sample.

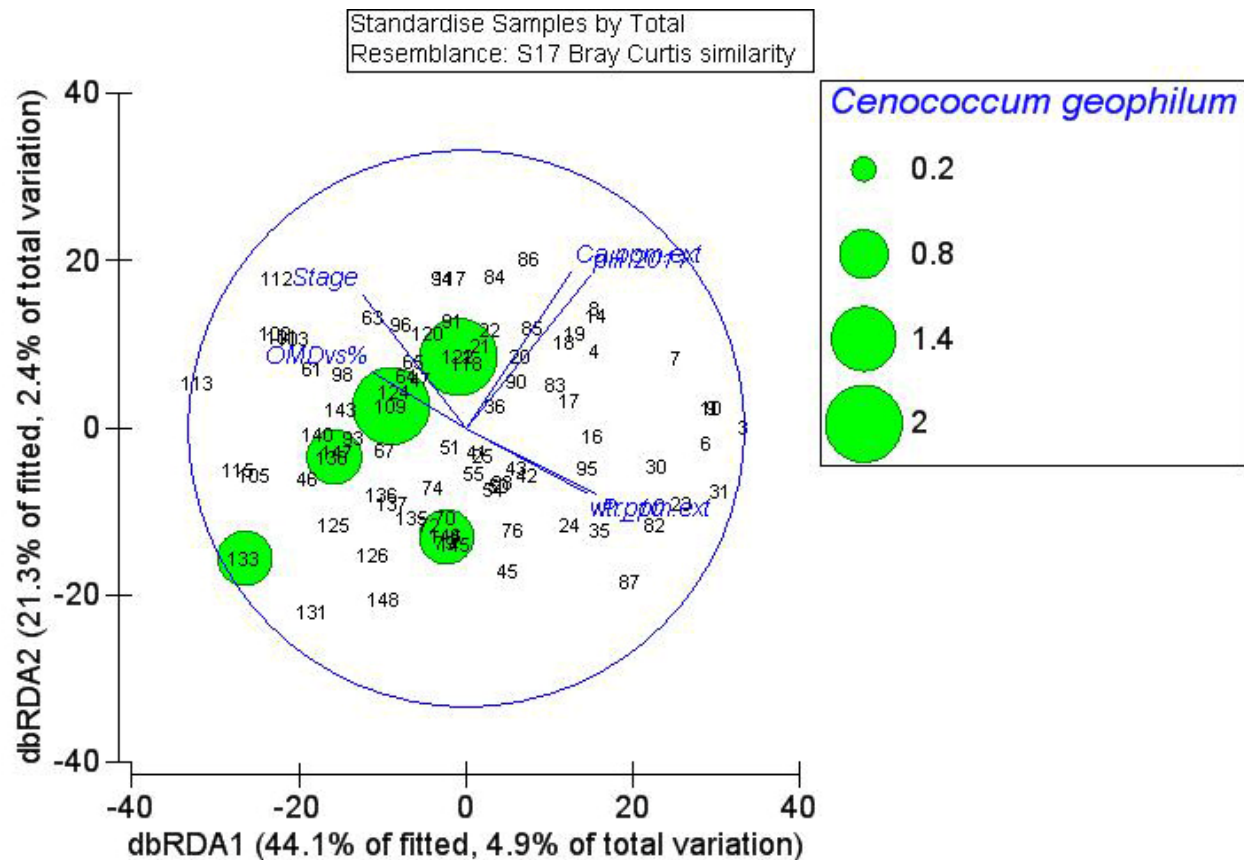


Figure 1-9 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both species level datasets (All species and species occurring in $>5\%$ of samples and the distribution of *Cenococcum geophilum* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Cenococcum geophilum* within that sample.

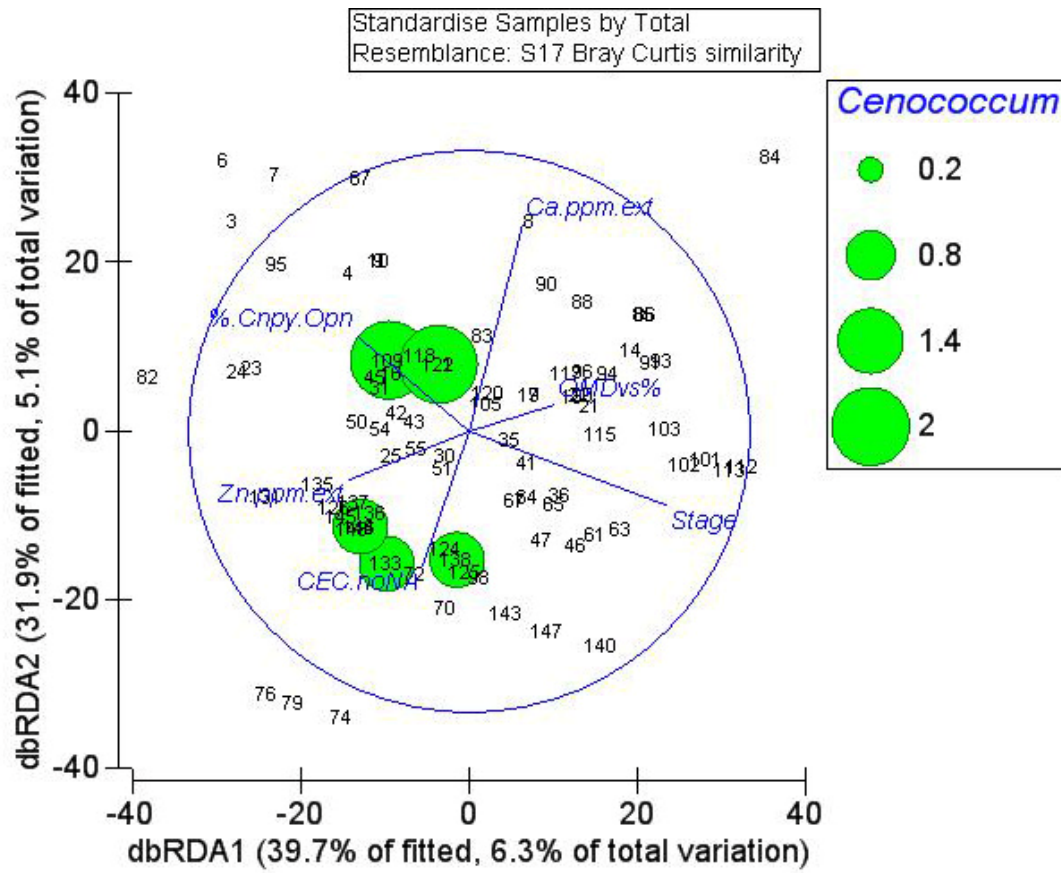


Figure 1-10 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both genus level datasets (All genera and genera occurring in $>5\%$ of samples and the distribution of *Cenococcum* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Cenococcum* within that sample.

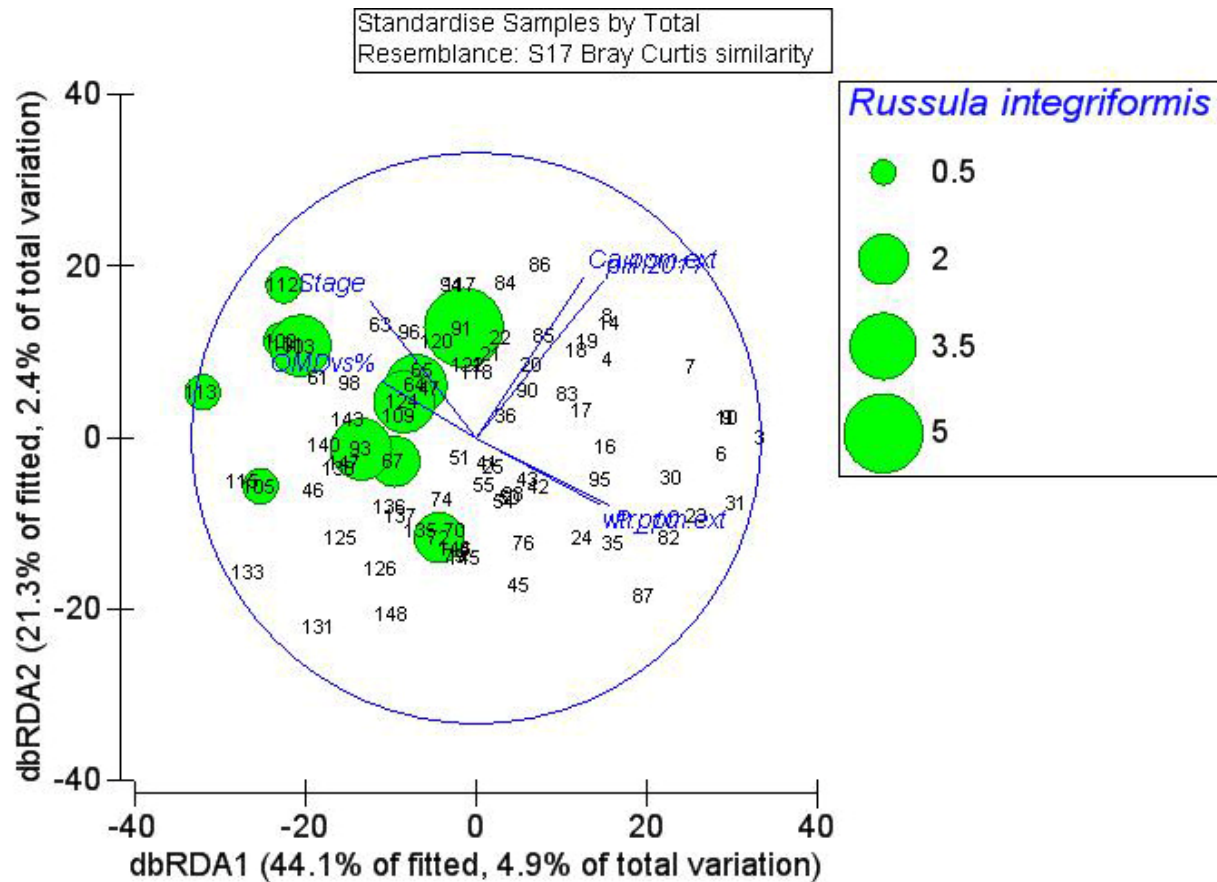


Figure 1-11 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both species level datasets (All species and species occurring in $>5\%$ of samples and the distribution of *Russula integriformis* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Russula integriformis* within that sample.

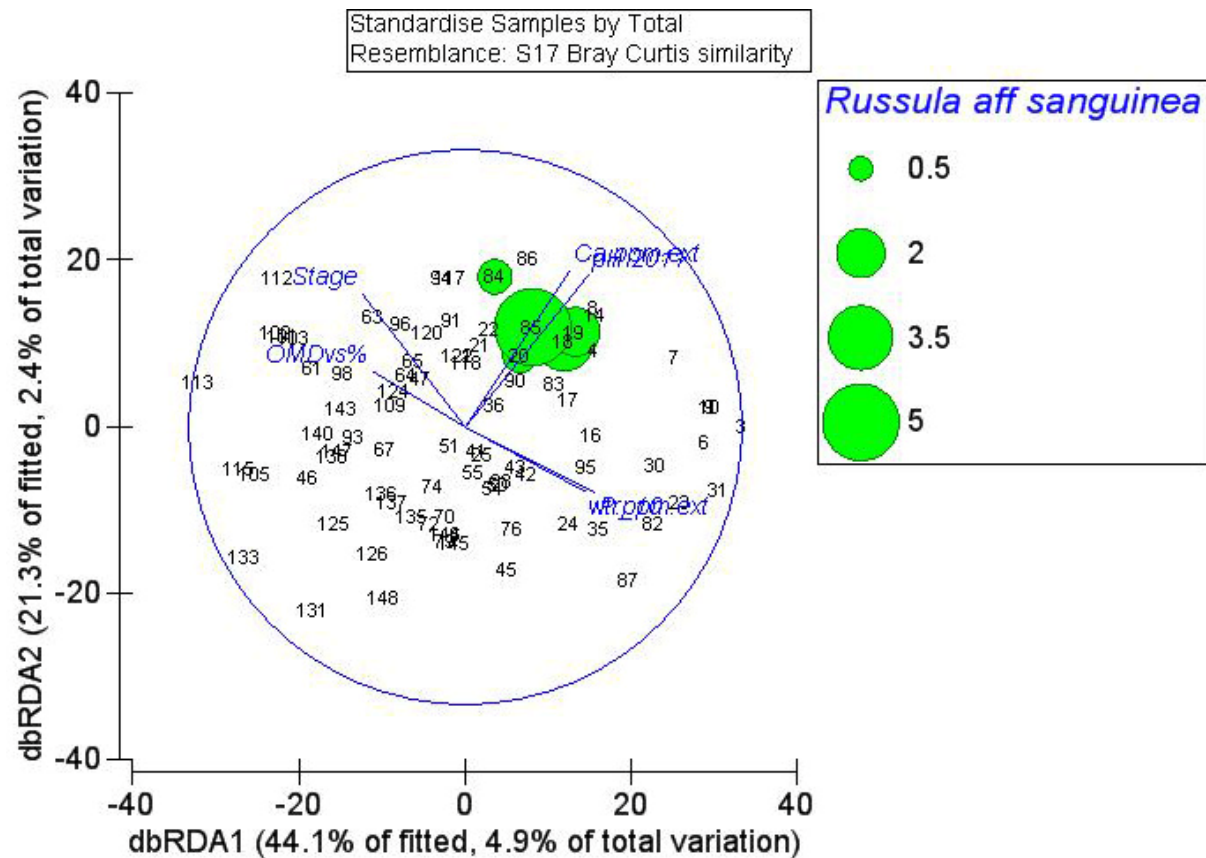


Figure 1-12 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both species level datasets (All species and species occurring in $>5\%$ of samples and the distribution of *Russula aff sanguinea* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Russula aff sanguinea* within that sample.

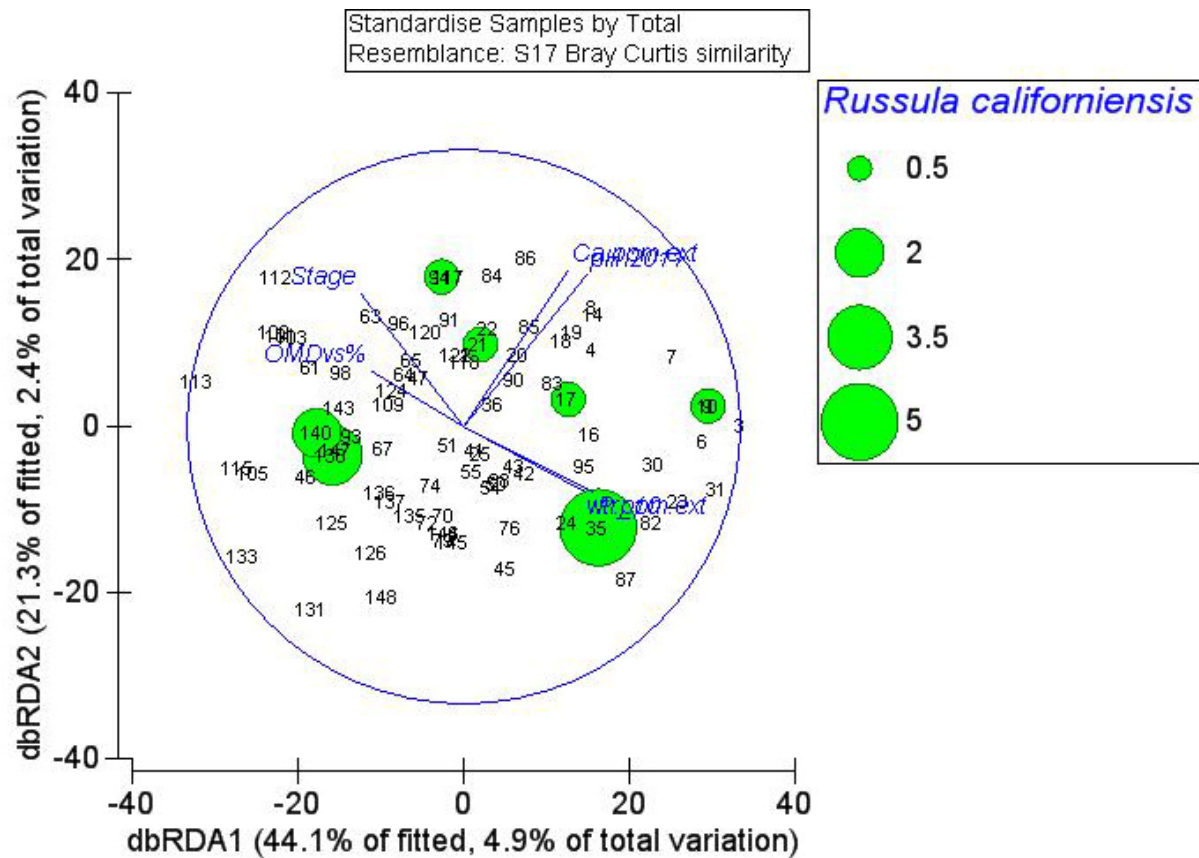


Figure 1-13 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both species level datasets (All species and species occurring in $>5\%$ of samples and the distribution of *Russula californiensis* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Russula californiensis* within that sample.

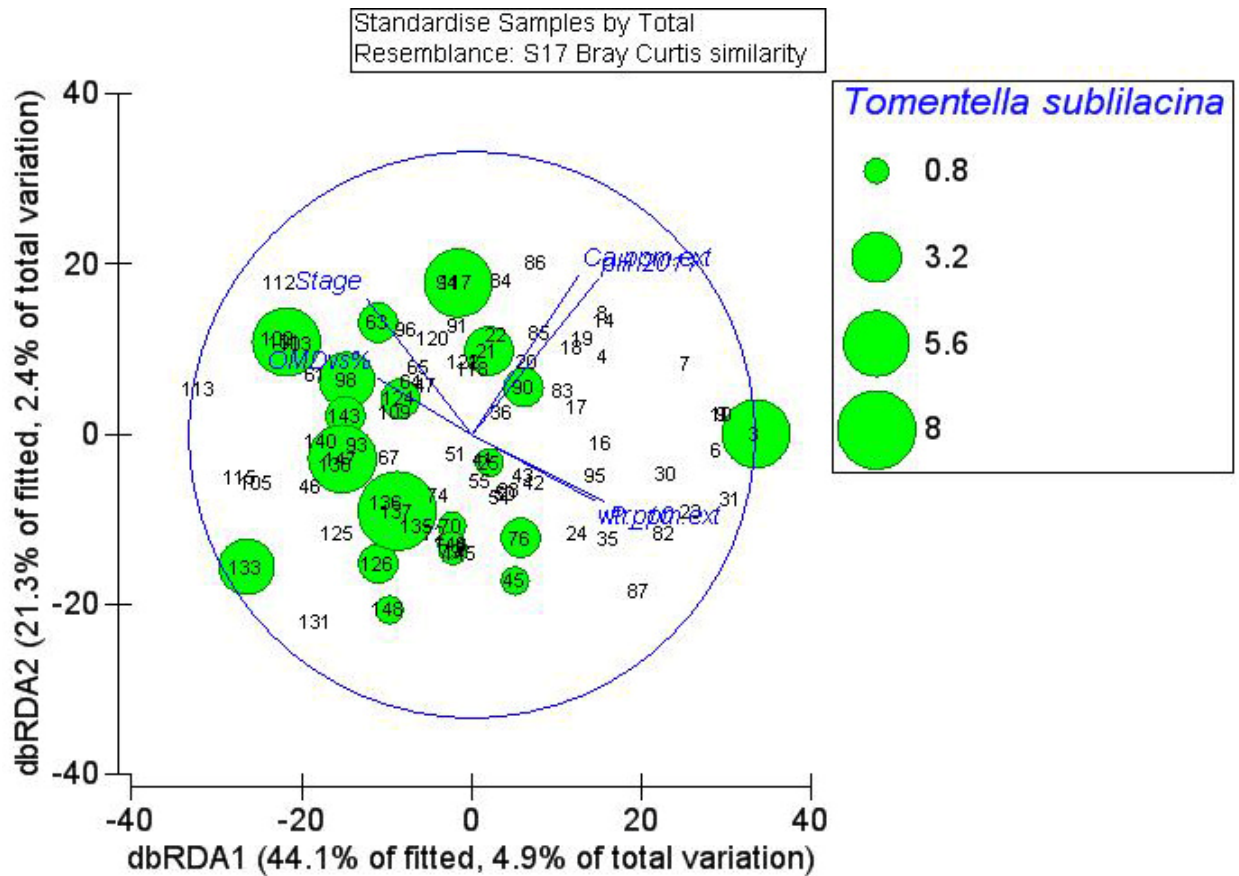


Figure 1-14 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both species level datasets (All species and species occurring in $>5\%$ of samples and the distribution of *Tomentella sublilacina* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Tomentella sublilacina* within that sample.

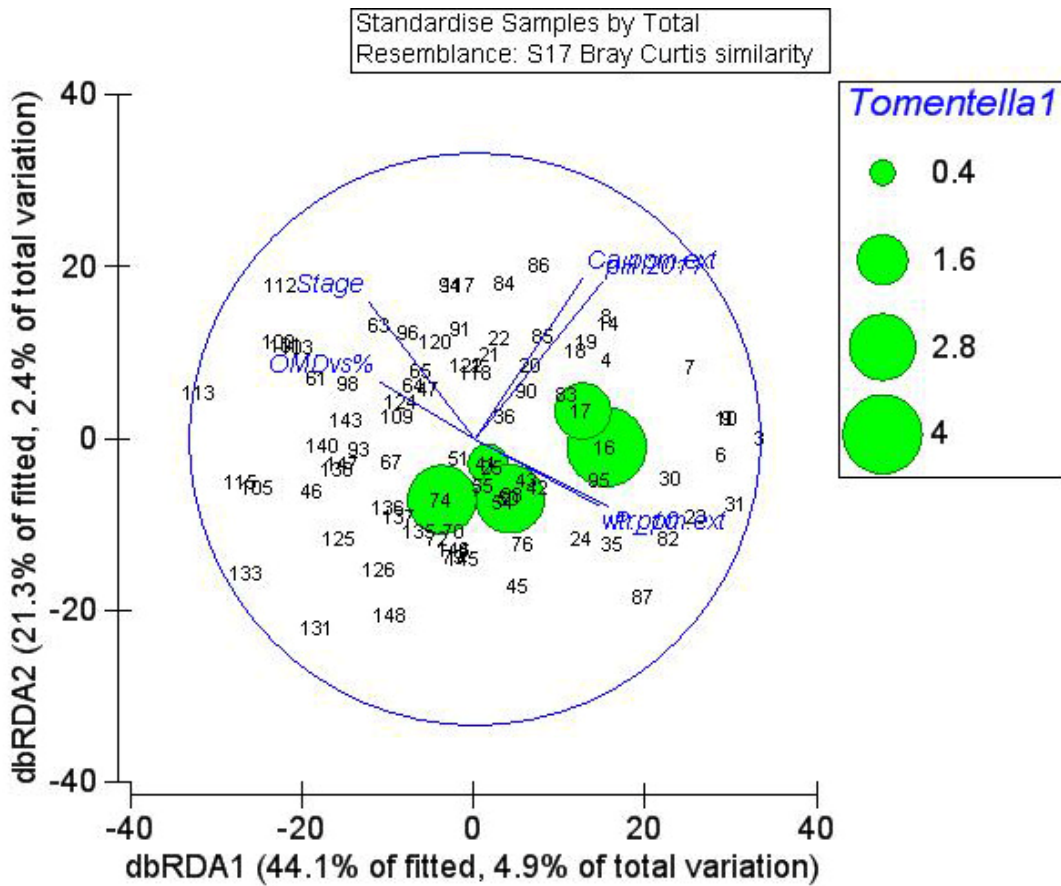


Figure 1-15 - Distance-based redundancy analysis incorporating variables with the highest relative variable importance scores (>0.3) for both species level datasets (All species and species occurring in $>5\%$ of samples and the distribution of *Tomentella1* in both plots within Cambria, California. Green circles represent presence and relative abundance of *Tomentella1* within that sample.

II: THE DEMOGRAPHY OF A NATIVE MONTEREY PINE (*PINUS RADIATA*) FOREST AND THE POTENTIAL CONSEQUENCES OF ECTOMYCORRHIZAL SUCCESSION ON FOREST GROWTH

INTRODUCTION

“The entire structure is neither tree root nor fungus alone but resembles the lichen thallus, a union of two different organisms into a single, morphological organ. It can be appropriately designated as a “fungus-root” or “mycorrhiza.””

-A.B. Frank “On the nutritional dependence of certain trees on root symbiosis with belowground fungi” (1885)

A key challenge in plant ecology is to gauge the strength and direction of factors that influence plant growth and structure plant populations. Since A.B. Frank’s 1885 publication hypothesizing the role and ubiquity of the ectomycorrhizal and plant symbiosis, ecologists have increasingly investigated the degree to and mechanisms by which mycorrhizal fungi affect the growth and mortality of plants (Frank 1885, 2005). Mycorrhizal fungi associate with nearly eighty percent of all plant species (Wang and Qiu 2006) and while they may fall along a continuum of effect on their plant hosts, they are generally regarded as beneficial (Johnson et al. 1997, Karst et al. 2008, Hoeksema et al. 2010).

Plant hosts may benefit from mycorrhizal colonization through both lower incidence and severity of diseases caused by plant pathogens (Sylvia 1983b) as well as provide some degree of drought tolerance at moderate levels of water stress (Boyd et al. 1986). Additionally, mycorrhizal fungi forage the soil for nutrients that may then be passed along to the host plant.

Ectomycorrhizal pines can receive up to 3.2 times more phosphorus and 1.8 times more nitrogen than their non-mycorrhizal counterparts (Bowen 1973). The benefits provided by mycorrhizal fungi consequently influence plant survival and growth; however, mycorrhizal species differ in their ability to perform these services and in their responses to plants (Allen and Boosalis 1983, Parke et al. 1983, van der Heijden et al. 1998, Sikes and Klironomos 2009).

Consequently, spatial or temporal variation in mycorrhizal community composition may drastically influence plant population dynamics and plant community structure.

Two mycorrhizal types, the arbuscular mycorrhizal and ectomycorrhizal (EM) fungi, have received the most attention in the ecological literature, largely because of the ubiquity and consequent ecological importance of their primary plant hosts, herbaceous and woody plants, respectively (Read 1991). Efforts to quantify the effects of variation in arbuscular mycorrhizal fungal composition upon plant populations have been largely successful, observing significant effects upon plant community structure including changes in plant diversity (van der Heijden et al. 1998, Hartnett and Wilson 1999, Klironomos et al. 2000, O'Connor et al. 2002), community composition (van der Heijden et al. 1998, Hartnett and Wilson 1999, O'Connor et al. 2002), and productivity (van der Heijden et al. 1998, Klironomos et al. 2000). In contrast, similar studies on ectomycorrhizal plant populations and communities have been stymied due to the long life span and delay in reproduction of many ectomycorrhizal hosts. Additionally, even shorter term experiments may be readily contaminated by non-target EM fungi. Hence, studies on the effects of EM fungal variation have necessarily remained in the realm of the short-term.

Land managers and conservation practitioners require information on the potential ramifications of variation in mycorrhizal fungal community composition for plant population dynamics to make informed and practical decisions to address applied problems. For this reason,

the effects of spatial and temporal variation in mycorrhizal fungal community composition on plant populations must be enumerated (Klironomos et al. 2011). Ideally, the long-term effects of variation in ectomycorrhizal fungal composition would be experimentally tested in the field; however, the logistics of effectively manipulating ectomycorrhizal fungal community composition for the periods necessary to study its long-term effects upon woody, perennial populations, make this approach unrealistic. Alternatively, by adjusting demographic data to reflect observed effects of ectomycorrhizal fungi in empirical short-term studies, models can be used to project and compare the long-term effects of ectomycorrhizal fungal community differences on various plant population parameters. Similarly, the incorporation of the observed demographic data into projection models can allow researchers to systematically perturb each demographic parameter at each stage/age individually, thereby generating data about the relative contribution of each demographic parameter of each stage/age to population growth. In generating these values, we can determine those phases of the perennial plant life-cycle where the effects of variation in ectomycorrhizal fungal community composition upon plant survival and growth are likely to have the greatest effect upon forest plant population. While this approach requires extrapolation of mycorrhizal effects, it is one step closer towards understanding the consequences of variations in ectomycorrhizal community composition for plant populations. The ability of modeling to estimate the long-term response of plant communities makes it well suited to address the effects of temporal variation of ectomycorrhizal fungal communities, or succession, upon plant populations.

The succession of ectomycorrhizal fungal communities has been demonstrated within a number of forest ecosystems (Fleming 1983, Fleming et al. 1985, Fox 1986, Visser 1995, Nara et al. 2003, Twieg et al. 2007, Yamashita et al. 2008). As the forest matures and plant roots

converge, vegetative growth of the mature forest ectomycorrhizal fungi creates a common mycorrhizal network (CMN) connecting the roots of these plants together. Seedlings may establish in the understory of these forests, connecting directly into these pre-existing CMNs, likely avoiding many of the carbon costs necessary to build their own (Simard and Durall 2004). Conversely, when seedlings in the forest understory are prevented from accessing CMNs, they are typically colonized by early-successional fungal species, whose total carbon cost must be paid by the seedlings themselves. Plants connected to the CMNs may create a “guild of mutual aid” making resources available to all regardless of identity or size (Perry et al. 1989, Egerton-Warburton et al. 2007). Resources that may be limiting to plant growth and survival, including water (Egerton-Warburton et al. 2007), nitrogen (He et al. 2003, He et al. 2005), and carbon (Simard et al. 1997) have been shown to move from plant to plant through CMNs; however, showing that CMNs affect the growth and survival of competing plants has proven difficult.

A recent manipulative experiment conducted in the native and mature Monterey pine (*Pinus radiata* D. Don) forest demonstrated a large, positive effect of CMNs on seedling survival. At the end of the two-year study, Booth and Hoeksema (2010) calculated a negative competitive effect of mature plant roots on seedling survival of -65% and a CMN mitigation of seedling mortality of +56%. This positive CMN effect on seedling survivorship may or may not have significant consequences for demographics of the forest, i.e. forest plant structure and population growth rates.

My study had three objectives. First, to summarize demographic information representative of the monodominant mixed-size class stands of Monterey pine in Cambria, California. Second, to understand the potential consequences of ectomycorrhizal succession (and the consequent formation of CMNs) for forest structure and population growth rates by

comparing model estimates of observed demographic growth rates to hypothetical “non-CMN” forests. I hypothesized that the large differences in seedling mortality (-56%) between the observed and hypothetical non-CMN models would translate to significant differences between model estimates of the finite rate of growth, λ . Third, to identify those demographic parameters most influential upon population growth rates so future studies may seek to address the relationships between mycorrhizal ecology and these phases in the life cycle of long-lived, perennial plants.

METHODS

Field Site

Cambria, California is host to the southernmost of three native California stands (35° 32' 25.21" N 121° 05' 29.57" W) of Monterey pine (*Pinus radiata*), covering c. 1400 hectares. The Kenneth S. Norris Rancho Marino Reserve (KNRMR) is located at the southern reaches of the Cambria stand and home to approximately 91 hectares of native Monterey pine forest. The KNRMR receives c. 460 mm of annual precipitation on average, most during the winter months, at which time most seedling recruitment and plant growth occur (Rogers 2002). Average temperatures range between 9° C and 18° C. The forest soil consists mainly of deep sandy loam (Carpenter and Storie 1933) which is low in organic matter (8 -15 cm) and easily erodes. The sandy soils and low summer precipitation are indicative that water and soil nutrients are the factors most limiting to plant growth and/or survival (Booth and Hoeksema 2010).

Monterey pine is a short-lived species, with a mean generation time of eighty or ninety years, infrequently living beyond one hundred and fifty years. Cambria Monterey pine average 30-37 m tall, may root as deep as soil or clay permit (~1.7 m in deep soil), and have extensive

lateral spread (9 – 12 meters) (Roy 1966). Additionally, Monterey pine are semi-serotinous such that fire stimulates massive seedling recruitment though cones also open and release seeds in response to seasonally warm, dry weather, so seedling establishment is common in the forest understory. Due in large part to encroaching urban and recreational development, the historic fire regimes of the California Monterey pine forests have been severely altered, the few occurring presently likely caused by accident or the occasional lightning strike (Rogers 2002). This shift from the natural fire regime within KNRMR in Cambria may be particularly stark, with no major fires occurring since at least 1940 (nearly a full generation for this species), and only a few small fires (~ 1 hectare) (Canestro Pers. Comm.). Therefore, the importance of the less prolific, but more constant and gradual seed rain from the semi-serotinous cones has shifted in its relative importance to the maintenance of these stands.

Monterey pine is the dominant tree species with only one other hardwood associate: Coast Live Oak (*Quercus agrifolia*). In moist soils, common understory plants are bracken (*Pteridium spp.*), California blackberry (*Rubus ursinus*), and poison-oak (*Toxicodendron diversilobum*) while drier sites include coast sagebrush (*Artemisia californica*), coyotebrush (*Baccharis pilularis*), and bush monkeyflower (*Mimulus aurantiacus*). Grasses along the edges of the forest include California oat grass (*Danthonia californica*), Blue wild rye (*Elymus glaucus*), and Purple needle grass (*Nasella pulchra*) (Canestro Pers. Comm.).

Collection of demographic data and data management

The demographic data used to determine size classes and to parameterize estimates of growth, survival, and mortality were collected from plots in two stand types representative of the Cambria forests, one a monodominant mixed size class stand of Monterey pine (93 m x 10 m,

MSC), and the other a stand of relatively equal proportions of coast live oak and Monterey pine (93 m x 20 m, OP) (Figure 2-1). Within each plot, the height of trees ranging from 1.4 – 3 meters tall was recorded, as was the diameter at breast height (1.40 meters, DBH) for all trees 1.4 meters tall or greater. In addition, height measurements of all trees shorter than 1.4 meters were recorded in a three-meter strip adjacent to the MSC plot and within five-meter strips on each side of the OP plot. Each tree was tagged and mapped along the plot for repeated annual measurements. DBH of all trunks was recorded when multiple trunks of an individual grew to 1.4 meters tall. Estimates of cone number, signs of disease, and mortality were recorded for all censused pines. When mortality was recorded, trees were reassessed for two additional census periods to ensure the tree was, in fact, dead. Full sampling efforts occurred in 2002, 2003, 2004, 2007, and 2009. An additional sampling bout in which only seedling recruitment was measured occurred in 2005.

Sources of data for matrices

Biological information chosen for inclusion in a Lefkovitch size class matrix model was collected largely from the monodominant MSC plot. To improve accuracy regarding growth and mortality data reflective of trees greater than 6.5 cm DBH, I incorporated data for those trees from both the MSC and OP stands. I chose these demographic data specifically since those two plots most accurately represent the sites in which Booth and Hoeksema (2010) conducted their study determining the relationship of CMNs and seedling survivorship rates in a native Monterey pine forest understory.

To accurately gauge the effects of CMNs on long-term population growth rates and stable size class structure, creating early size classes representative of one-year-old recruits and two-

year-old individuals was crucial. Therefore, in delimiting size classes for these aged individuals, I only used data collected in consecutive years ('02 – '03, '03 – '04, and '04 – '05) in which I knew the age of one- and two-year old plants.

To utilize the demographic data of plants ≥ 140 cm tall to the fullest extent, where census periods were separated by a year or more of no data collection, growth was assumed to have occurred evenly over the intervening years. In these intervening years of no census, if mortality occurred, I had no knowledge if that death occurred in year J or J+1. Although mortality data of individuals ≥ 140 cm in height in successive years suggests the probability of death to be more likely to occur in year J + 1, I constructed two matrices from the original data, one in which all unknown mortality occurred in year J and the other in which all death occurred in year J + 1. Estimated λ s of the resultant matrix projections of these two extremes did not differ appreciably and so only the results from the latter scenario are reported.

Finally, although the data showed instances in which DBH actually contracted slightly from one year to the next, for purposes of modeling, no trees were allowed to transition to smaller size classes. Additionally, for trees with multiple stems, I calculated a total DBH following the calculation:

$$\text{EQ 1: Total DBH} = \sqrt{\text{DBH}_a^2 + \text{DBH}_b^2 + \text{DBH}_c^2 + \text{DBH}_n^2} \dots$$

where DBH_{a-n} are DBH measurements on multiple stems a-n.

Size Class Determination and Matrix Construction

Size classes defined by high mortality rates

One of the primary objectives of this chapter was to compare the growth rates and size class structure between the observed demographic data and an adjusted dataset in which the

CMN effect (an increase in seedling survival rate after two years of 56%) observed by Booth and Hoeksema (2010) was removed. The boundaries of size classes one and two, representing first year recruits and second year individuals, were determined from the mean heights of individuals of that age \pm two standard deviations. The upper bound of the 1st size class overlapped the lower of the second size class slightly, but the second size class had an upward-skewed distribution, so I set the upper bounds of the first class as the lower bounds of the second.

Often, one and two year olds were observed in the data to remain in their respective size class for additional years; therefore, the 1st size class included 2-,3-, and 4-year-old individuals, and the 2nd size class included 3-,4-,5-, and 6-year-old individuals.” Theoretically, a small proportion of the first and second size class individuals could remain in these size classes indefinitely, but it would be impractical to create a matrix model to account for this possibility, so I set a 0.005 probability limit to stop including additional age matrices within the 1st or 2nd size classes. In this model, the final age a plant could be and still remain in a size class was determined by multiplying the probability of stasis in a size class by itself until this probability was < 0.005 .

Creation of discrete size classes required a concise point at which I was to start classifying individuals into size classes based upon DBH, rather than height. Hence, the fourth size class was determined by taking the mean height \pm two SD of individuals growing more than 140 cm tall into the next year. The third size class included those individuals falling in size between size classes 2 and 4. Caswell (2000) suggests choosing size class groupings that correlate strongly with some biologically relevant life characteristic (e.g. probability of death, growth rate, reproductive effort). Additional support was lent for the first four size classes as the likelihood of mortality to individuals from year to year differed strongly between these groups

(Table 2-1). Due to occasional gaps between sampling years, I based the probability of death of these earlier classes on individuals for which I had data in successive years (2002-2003, 2003-2004).

Size classes defined by cone production

Cone production correlated strongly with DBH (0.856, $p < 0.001$); therefore, cone production was used as the defining feature of the mature size classes. Two reproductive groups were defined by determining the two highest DBH quartiles from those cone-producing trees ($DBH \geq 55$ cm and $42 \leq DBH < 55$ cm). Due to large disparities between minimum and maximum cone production of trees $DBH \geq 55.0$ (10 – 301 cones), this group was further delineated by median cone production into two groups, 55 – 61.4 and > 61.4 cm DBH. Fecundity estimates were calculated under the assumption that contribution to recruitment was equal to cone production of each size class relative to total cone production of the documented population, divided by the mean annual number of individuals with each size class, multiplied by the mean annual recruitment number:

EQ 2: *Recruits attributable to size class S individual in year J + 1 = ((Size class S cone production in year J / total population cone production in year J) / N individuals in size class S)*

** N recruits in year J + 1*

Size classes defined using Vandermeer-Moloney algorithm

Individuals > 140 cm tall but less than 42 cm DBH showed less distinctive correlations with biologically relevant life characteristics. To partition these individuals into size classes for the model, I employed the Vandermeer-Moloney algorithm as suggested by Caswell (2000, pg.

169) and outlined by Moloney (1986). Vandermeer (1978) developed this algorithm to reduce two kinds of error involved in choosing size classes, standard error (SE) and distribution error (DE). The SE occurs as size classes become too narrow, reducing the number of individuals within each size class, creating error in the estimates of biological parameters used in the model. In contrast, DE is caused as size classes become increasingly large and growth information of individuals is lost. Vandermeer's solution to this problem of conflicting errors was to develop an algorithm to pinpoint size classes where the sum of these errors is minimized. Therefore, the choice of these intermediate size classes, with no clear biological division, was determined by choosing increments that reduced the summed value of SE and DE.

Modeling

Modeling of observed forest data

All relevant biological parameters (stasis, growth, mortality, recruitment contribution; Table 2-1, Figure 2-2, Figure 2-3) from the observed demographic data were entered into a matrix, and multiplied by an arbitrary starting point of 1 individual in each size class, to estimate the population growth rate by calculating the finite rate of increase, λ , and determining the stable size class structure of the population. I used a modified form of the traditional bootstrapping technique (described below) to calculate 95% confidence intervals for λ .

Regardless of the initial population size in each size class (which is multiplied by the estimated demographic parameters entered into the matrix), the population will always converge on the same stable size class structure and value for λ ; however, the number of iterations required to converge upon these values will vary. If observed data for current size class structure is used for the initial population sizes, then the number of iterations required for convergence may be

indicative of the degree to which the current structure deviates from the predicted stable size class structure. Therefore, using the observed size class distributions of the 2009 demographic data as a multiplicative starting point against the observed data matrix, I calculated the number of iterations (years) required before the proportional distribution of every size class was within one percent of the stable size class structure.

Elasticity Analysis

Elasticity analysis is conducted by perturbing each value within the projection matrix equally, one at a time, and comparing the relative effect these changes have on the resultant λ (de Kroon et al. 1986, Silvertown and Charlesworth 2001). In doing so, I sought to determine those size classes of the Monterey pine tree life cycle where small changes in demographic parameters are most likely to affect population growth rates. It is likely these size classes where differences in plant responses to variation in ectomycorrhizal fungal community composition will have the greatest effects upon plant population dynamics and community structure (Caswell 2000).

Elasticity values sum to one to allow for comparisons between demographic parameters; however, it is known that larger/older trees grow more slowly than smaller or younger trees (Gower et al. 1996). Additionally, the final size class in a projection matrix has no additional size class to grow into meaning that the calculation of the stasis parameter for this size class includes demographic information applicable to growth and stasis. By summing stasis and growth elasticities, and dividing by an estimated maximum residence time (MRT) of each size class, we may calculate an adjusted survivorship elasticity. This metric is a proxy for the elasticity of annual survivorship and is, perhaps, a more accurate descriptor of the importance for survivorship in a size class. This adjusted annual survivorship elasticity is only comparable

between adjusted annual survivorship elasticities of other size classes or to the unadjusted survivorship elasticity values and not to the elasticities of fecundity, stasis, or growth. Although this treatment of elasticity data is rarely conducted upon stasis and growth information in other demographic studies, I felt it was important to do so in order to gain additional perspective on how annual survivorship within size classes potentially affects population growth rates and juxtapose this information with the elasticity values for the unadjusted survivorship values.

Modeling of the hypothetical, non-CMN forest: adjusted models

To investigate the long-term effect of ectomycorrhizal fungal succession and consequent development of CMNs upon forest populations, I developed two models that remove the CMN reduction of seedling mortality observed by Booth and Hoeksema (2010). In their paper, mortality was reduced by 56% at the end of the two-year study. In fact, this net two-year reduction in mortality was distributed unevenly over the study, with a 14.4 % reduction in the first year, and a 26.6% reduction in mortality for the individuals remaining in the study after the first year. It is possible that these reductions in mortality were age or size specific and so additional age classes within size classes 1 and 2 were incorporated, and CMN-related survival benefits were removed from those age classes in two different ways:

Model A: The first and more conservative of these two models removes CMN-related survival benefits only from the 1st (-14.4%) and 2nd year (-26.6%) age classes in size class 1, and from the 2nd year age class in size class 2 (-26.6%), but not from the other additional age classes in size classes 1 and 2. This model aims to understand the ramifications of the Booth and Hoeksema (2010) observed CMN-related survivorship benefits on population growth rates, assuming that their results apply to one and two year olds, regardless of the size of these plants.

Model B: The second and more liberal of these two models removed the CMN-related survivorship benefits found by Booth and Hoeksema (2010) from all age classes in size classes 1 and 2. Therefore, all size class 1 ages were attributed the -14.4% survival reduction and all size class 2 ages received the -26.6% survival reduction. This model contrasts the prior, more conservative model by assuming that CMN-related survival benefits are size specific, but not age specific. As stable size class distributions of these two models did not differ noticeably, I limit comparisons of stable size class distributions to Model B. However, for purposes of discussing the effects of CMN-related survivorship reductions on λ , we discuss both Models A and B.

The A and B models are the most conservative model estimates of the effect of CMN removal on forest demography, since they assume that CMNs only affect survival of either the one- and two-year olds or the first and second size classes. While Booth and Hoeksema (2010) ended their study after two years, it is possible that these reductions in mortality by CMNs could persist further into the life cycle of the trees. Therefore, I sought to understand the potential life-long influences of CMNs upon forest stable size class structure and population growth rates, λ . To do so, I developed an additional 15 models, each removing the second-year CMN mortality reduction of 26.6% from additional and progressively later size classes in the tree life cycle. The final two models, one removing the CMN survivorship reduction from all size classes but the last, and the other removing the CMN survivorship reduction from all size classes, will be hereafter referred to as X and Y, respectively.

Modified bootstrapping for 95% confidence intervals

For all datasets, real or altered, I modified the traditional bootstrapping technique so that randomized sampling with replacement of each size class, except the final size class, would

continue until at least one instance of growth into the next size class was observed in the new bootstrapped dataset. Bootstrapping of the final size class did not use this contingency, but I ensured that at least one death was recorded. Without these modifications, the calculations of λ would not be possible for model iterations in which size classes 0 – 16 did not have one member moving onto the next size class and would, similarly, fail to stabilize if death did not occur in the last size class. All bootstrapping to calculate 95% confidence intervals for the finite rate of increase, λ , were derived from 50000 iterations of the real or modified growth and mortality data, while size class fecundity was held constant.

Software

PopTools version 3.2.3 was used to calculate the finite rate of increase, λ , as well as stable size class structure for each model and elasticity analysis for the observed demographic data (Hoeksema et al. 2010, Hood 2010). Similarly, PopTools was used to calculate the MRT within each size class. R statistics was used to calculate λ and 95% confidence intervals for all matrix models. Additionally, R statistics was used to calculate intermediate size classes from the observed demographic data using the Vandermeer-Moloney algorithm (Moloney 1986). The finite rate of increase, λ , was calculated using both PopTools and R statistics but the two calculations did not differ appreciably, so I report only the results from R statistics.

RESULTS

In total, I specified 17 size classes characterized by growth rates, mortality, and reproduction (Table 2-1, Figure 2-2, Figure 2-3). Probability estimates of stasis, growth, mortality, and contribution of recruits in the following year per individual of each size class were

calculated from demographic data collected on 492 and 341 trees, for size classes 1–4 and size classes 5–17, respectively.

Modeling of observed forest data

Modeling of the observed forest data estimated a ~ 2% yearly reduction in population size ($\lambda = 0.9822$; Table 2-2) of the mixed-size class *Pinus radiata* populations in Cambria, California at KNRMR. This result assumes fixed vital and reproductive rates of the population. The projected stable size class structure resembled an inverse-J curve (Figure 2-4, Figure 2-5). In this stable size class projection, the large majority of plants (~82%) reside within the first size class; however, thereafter numbers of plants within size classes dramatically and progressively decline since the large majority of size class 1 individuals do not survive into higher size classes (Table 2-3, Figure 2-4, Figure 2-5). Additionally, a large proportion of recruits are maintained in the population due to reproduction by fecund mature size classes. Using the 2009 demographic data to determine an estimated length of time before convergence upon the predicted stable size class structure yielded a 50-year time span.

Elasticity Analysis

Elasticity values for growth out of or stasis within the first and second size classes were appreciably lower than those of the following size classes, and orders of magnitude less than those of some later size classes. Elasticity analysis suggests that population growth rate is most influenced by the stasis of later size classes, even more so than reproduction within or growth out of these same later size classes, and of these later size classes, stasis within the final size class appears to be most influential of all. Changes to values of fecundity had little relative influence

upon population growth; however, changes to later size class fecundity values had more influence than those associated with earlier size classes (Table 2-4).

Calculation of elasticities of unadjusted size class survivorship and the estimated adjusted annual survivorship revealed large relative differences within and between these categories. The unadjusted survivorship elasticities indicate a much greater potential for changes in survivorship to later size classes, specifically size classes 14 – 17, to influence population growth rates (2 – 20 times greater). In stark contrast, estimates of elasticity for adjusted annual survivorship of size classes suggest that survivorship is relatively important throughout most of the life cycle of this species (Size classes 3 – 17); however, seedling elasticities (Size classes 1 and 2) for estimated annual survivorship are still relatively low in comparison those later size classes (Table 2-4).

Modeling of the hypothetical, non-CMN forest: adjusted models

Model projections of reduced survivorship only for one and two year-old plants within either size class one or two (Model A), reflective of the findings by Booth and Hoeksema (2010), had no significant effect upon population growth rates of the population (Table 2-2, Figure 2-6). Similarly, the model projection of these survivorship reductions upon all age classes of the first and second size classes (Model B) was not significantly different from the observed population projections λ or Model A (Table 2-2, Figure 2-6). Similarly, the iterative application of the reduction in survivorship (-26.6) to models including higher size classes (up to size class 16) were not significantly different from the λ of the observed projection model. In contrast, I observed a dramatic and significant decrease in λ when this reduction in survivorship was applied to the entire life cycle of this species (Table 2-2, Figure 2-6).

Calculations of stable size class structure for the adjusted models showed a relatively small, though noticeable shift from that determined for the observed forest data. As the reduction of survivorship was incrementally attributed to higher size classes, a greater proportion of the population resides in size class one and in later size classes. These incremental increases continued with progressive reduction of survivorship within each size class until the survivorship reduction was attributed to the entire tree life cycle, whereby the proportion of the population represented by each size class returned to similar levels of those seen in the observed demographic data. As a result, the projected stable size class structure of the forest without any CMN effects on mortality was similar to the projected size class structure using observed demographic data (Figure 2-4).

DISCUSSION

The demographic work conducted in this study indicates that this stand of Monterey pine has a negative growth rate, and hence if this growth trend continues, the population will go extinct. Additionally, model comparisons indicate that EM fungal community succession and consequent development of late-stage CMNs that have been shown to positively influence seedling survivorship (Booth and Hoeksema 2010) may not significantly influence growth rates or stable size class structure of the Monterey pine population. If, however, these CMN effects are not limited to seedlings, but instead continue to influence survivorship throughout the tree's life cycle, the successional development of these fungal networks may be extremely important for survival of Monterey pine and other forests where stands display higher levels of seedlings recruitment or survival (e.g. those that have been recently disturbed).

Finally, these results are derived from a deterministic model utilizing available demographic data. The differences between the projected stable size class structure and that observed in the actual forest suggest that this population experiences or has experienced a variable environment not captured in the demographic data collected since 2002. Continued monitoring of these stands may reveal the source of this environmental variability and lead to the subsequent development of a stochastic model that projects a population structure that more closely resembles that observed in 2009. Conversely, the frequency or magnitude of past environmental variation may not be as great as it has been historically, for instance through active suppression of the natural fire regime. Continued monitoring of these stands may help to confirm the results of this study by failing to observe great swings in forest demographics.

Demography of a native Monterey pine stand

There have been numerous studies conducted on the demography of Monterey pine; however, many of these are limited to exotic stands (Acuna et al. 2010, Mosquera-Losada et al. 2010), a large percentage of which are specific to plantations in Australia and New Zealand (e.g. Dzierzon and Mason 2006, Haywood 2009), with a growing number addressing the increasing incidence of invasion posed by Monterey pine in non-native habitat (Williams and Wardle 2007, Baker and Murray 2010, Gomez et al. 2011). In contrast, relatively few studies have been conducted within the native range of *Pinus radiata*, even fewer of which have investigated population structure or growth (but see White 1999, Stephens et al. 2004, Piirto and Valkonen 2005, O'brien et al. 2007). None, to my knowledge, have tracked a native stand for consecutive years for longer-term demographic purposes.

Estimated population growth rates of the observed stand were significantly lower than one ($\lambda = 0.9822$; 95% CI = 0.9684, 0.9985), suggesting that if growth, mortality, and reproduction estimates remain constant, the population will eventually be extirpated. It is important to note that these coastal Monterey pine populations reside in environmentally variable habitats and that recruitment events post-fire may be significant. As indicated by the 2009 demographic data (Table 2-3, Figure 2-6), these stands are of mixed-size class structure, a feature common to forests experiencing low severity fire regimes (Agee 1998) that do not cause a massive release of the aerial, semi-closed cone seed-bank, or one in which the fire regime has been largely removed from the system. In lieu of its natural fire regime, Monterey pine is known to release some amount of its canopy stored seeds in response to dryer environmental conditions (Roy 1966); however, increasing forest floor detritus or increased canopy shading may limit the germination of these seeds and therefore, the number of seedlings may be insufficient to perpetuate the population (Stephens et al. 2004).

The size class distribution observed during the 2009 census differs noticeably from that of the projected estimates of stable size class distribution. A spike in the demographic distribution of size classes 4 and 6, versus the typical J-curve distribution common to mixed-age forests (Peng 2000) seen in the stable size class estimate (Table 2-3, Figure 2-6), suggests a degree of instability in the population, with a relatively strong cohort moving through the population. Estimates of the time to a stable size class structure of 50 years agree with this finding, indicating the relative degree to which the size class structure of the 2009 population differs from that calculated from the deterministic model. This 50-year estimate may be partially explained by the presence of these strong cohorts within the 2009 data, and the time frame it will take for them to pass through the population. Contrastingly, this estimate may be an indication

that the population is shifting from the historic natural fire-maintained size class structure to one in which fire has been removed from the system.

Common mycorrhizal network effects on forest demographics

My hypothesis that the large differences in seedling mortality (-56%) observed between seedlings with and without access to mycorrhizal networks in a field experiment (Booth and Hoeksema 2010) would translate to significant differences between model estimates of the finite rate of growth, λ , was not supported when considering only the first two size classes. The significant overlap between estimates of λ for the observed demographic data and models A and B suggests that the mortality reductions of Monterey pine seedlings caused by CMNs will not likely translate to long-term population level effects upon plant community dynamics or community structure (Table 2-2, Figure 2-6). In fact, out of the 17 hypothetical non-CMN models, no estimated λ s were statistically different from that estimated for the observed demographic data, except, that of the final model, Y, which removed CMN survival benefits from the entire life cycle of the Monterey pine. This result is at least partly in agreement with the elasticity analysis of the observed demographic data, which suggests that changes in mortality for smaller size classes will have little repercussion for growth rates of the population. These findings are consistent with the comparative study of 45 herbaceous and 21 woody plant elasticity values conducted by Silvertown et al. (1993) that found that growth and fecundity were significantly less important to the λ of woody plants than to herbaceous plant species and conversely, survival was of greater importance to woody species than herbaceous plants. Calculation of unadjusted survivorship agree with this study's findings; however, comparisons between elasticities of unadjusted survivorship and estimated annual survivorship for each size

class indicate a much greater relative role of seedling survivorship in influencing population growth rates. Still, the estimated annual survivorship elasticities suggest that changes to survivorship of later size classes will be more influential to population growth rates than similar changes to the youngest size classes of trees.

The final model, Y, estimated a drastically and significantly lower λ than that of any other model, including the observed demographic data (Table 2-2, Figure 2-6). It is somewhat surprising that the models including survival reductions for other later size classes including 14, 15, and 16 did not exhibit a significant shift in population growth rate, λ . Elasticity values for these size classes, specifically for stasis, were appreciably higher than other lower size classes and additively, much greater than that of size class 17. It is possible then that this final size class, lacking high mortality rates and predisposed to high reproductive rates (Table 2-1), would maintain an adequate number of recruits in the population to supply, over time, additional mature, fecund individuals to perpetuate the population. Comparisons of stable size class distributions between the observed model, model X, and model Y lends support for this idea whereby greater proportions of those late size classes, especially the last, maintain a proportionally greater amount of recruits in the population (Figure 2-4).

Model fidelity

The results presented here are reflective of the demographic data collected in this stand since 2002. The deterministic modeling approach taken here has been argued to be more accurate when there are five or fewer consecutive years of data (Doak et al. 2005), while stochastic models are preferred when enough years of data have been collected. This preference for stochastic models is true even more so within environments characterized by large climatic

fluctuations or for populations that experience frequent or semi-frequent disturbance. Monterey pine is a fire adapted species and so fire disturbance may result in greater seedling recruitment events that could drastically affect population growth rates. It should be noted that mortality estimates were excessive, with mortality of the first size class exceeding 80% (Table 2-1, Figure 2-2). Similarly, fecundity values were modest in comparison to those expected to be expected post-fire (Table 2-1, Figure 2-3). Roy (1966) observed increased recruitment and growth of Monterey pine in areas of bare, mineral soil and increased light, i.e. just those environmental characteristics one would expect to change following a minor or major forest fire (Stephens et al. 2004). Additionally, Fenton (1951) observed recruitment following a stand fire estimated at $\sim 900,000$ seedlings ha^{-1} . Although perhaps not entirely applicable due to its occurrence in a Monterey pine plantation, this observation highlights the importance for fire in recruitment of this species. Therefore, the calculation of elasticity values for a population experiencing periodic stimulation of seedling recruitment may differ dramatically from that observed in this study (Table 2-4). The reality however, is that fires are actively suppressed in the California stands of Monterey pine due to encroaching human development. Introduction of fire into these habitats could greatly increase recruitment and survivorship of seedlings, at least in the short term, which may increase population growth rates significantly.

The differences between the stable size class distribution and the observed size class distributions in 2009 suggest that these populations reside in a variable environment; however, the demographic data of the past nine years do not reflect this variability (Figure 2-5). The model estimates presented above may then be a forecast of how these populations will respond in the long term with continued fire suppression or limited environmental variability. Additional

demographic work within this, and other populations, could provide more accurate model estimates of population growth rates and validation or refutation of these results.

CONCLUSION

There is a growing body of literature suggesting that variation in plant growth and survivorship is strongly influenced by ectomycorrhizal fungal community composition (e.g. Parke et al. 1983, Booth 2004, Booth and Hoeksema 2010); however, these studies have (perhaps out of necessity) all focused on how these compositional changes affect young, immature woody-plants. The modeling approach that I have employed in this study suggests that while CMNs may strongly influence the mortality rates of Monterey pine seedlings (Booth and Hoeksema 2010), the long-lasting influence of these effects on populations of Monterey pines, at least measured by the metrics of population growth rate and stable size class structure, may be modest (Table 2-2, Figure 5-2, Figure 4). If this is true, the temporal variation of EM fungal communities observed during succession that leads to the development of these CMNs may be of little consequence for forest populations. If, however, the observed benefit of CMNs for plant survivorship endures throughout the life cycle of Monterey pine, EM fungal succession could drastically influence plant population dynamics in the long term in these forests. Importantly, relatively small differences in survivorship or growth for later size classes in the woody plants life cycle may strongly influence population growth rates (Table 2-4).

It is, perhaps, a cruel irony for the ectomycorrhizologist that those life cycle stages most strongly implicated in influencing woody plant population growth rates are the most difficult, logistically, to quantify in their response to variation in EM fungal community composition. This result, however, should not limit investigation into this realm as science has consistently

bypassed obstacles that once seemed insurmountable. In order to understand the potential for variation in ectomycorrhizal communities to influence populations of woody, perennial plants, researchers must begin to construct studies that aim to uncover its effect upon the growth, survival, and fecundity at later stages of plant development. It may be necessary, in turn, to take a multifaceted approach to this problem by invoking a combination of methods including, but not limited to, long-term research sites and extrapolative modeling.

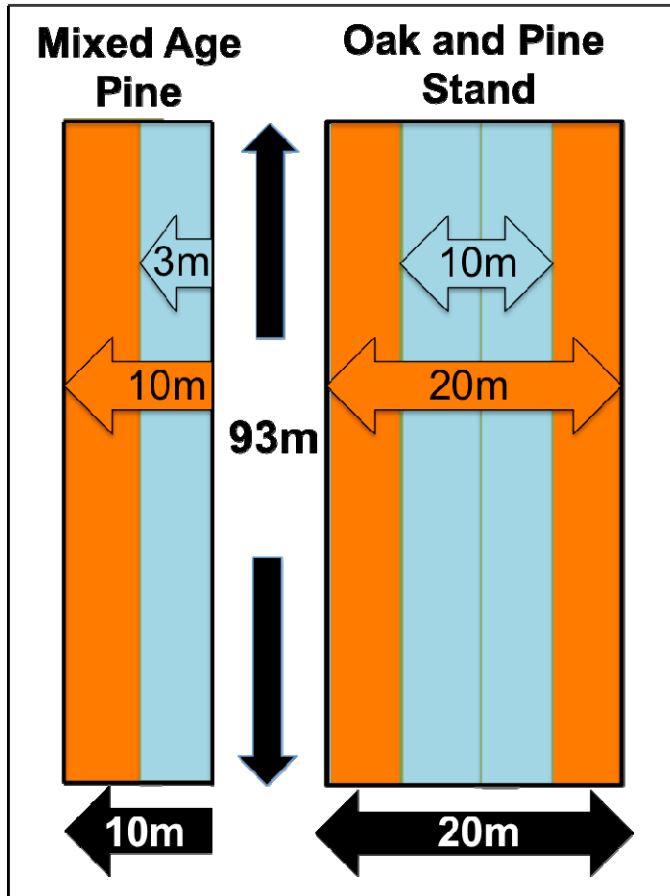


Figure 2-1: Diagram of forest plots in which data for the demographic matrix projections were collected. All data collected from the mixed age pine stand (MSC) was used for model estimates along with all data for trees 6.5 cm diameter at breast height or greater in the oak and pine stand (OP). Regions in blue represent those areas of the plots fully sampled for trees of all sizes. Regions in orange represent those areas of the plots where only trees ≥ 140 cm tall were sampled.

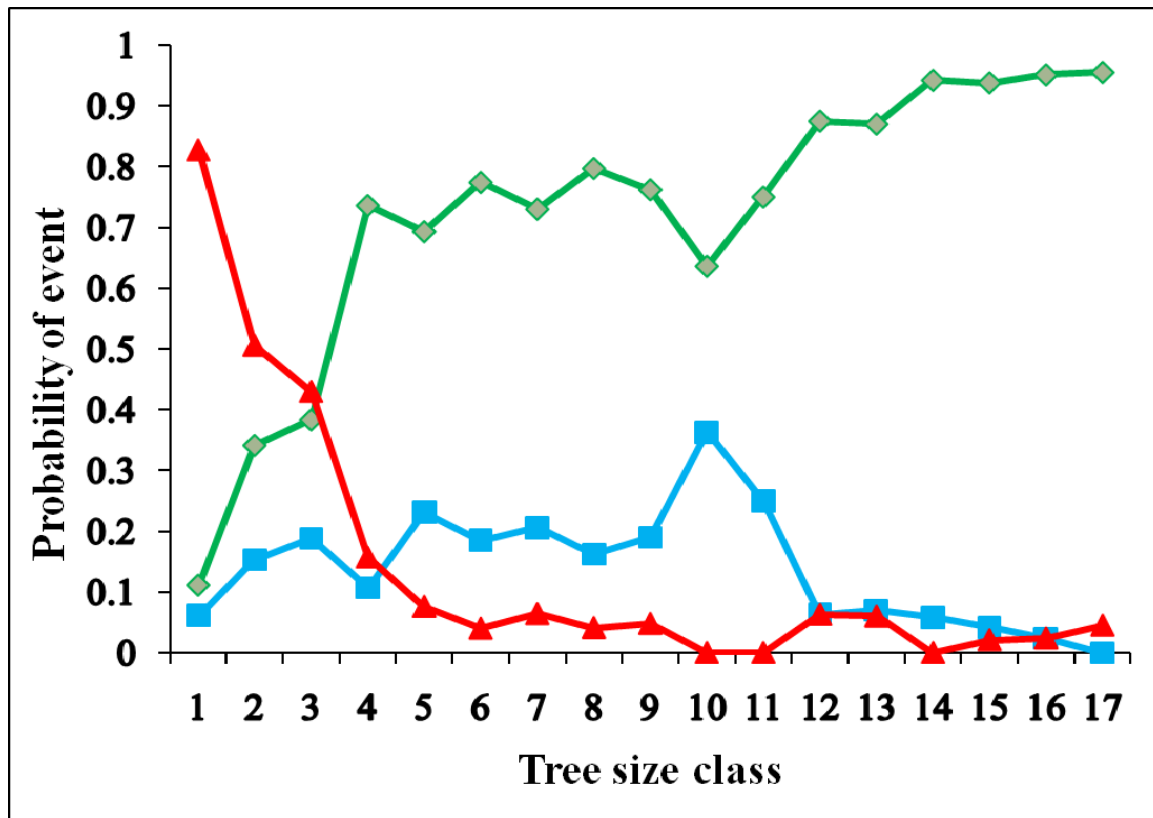


Figure 2-2: The average probability of a demographic event occurring for each size class for stasis (remaining in a stage into the next year, green), growth (growing into the next stage in the following year, blue), and mortality (dying in the present year, red).

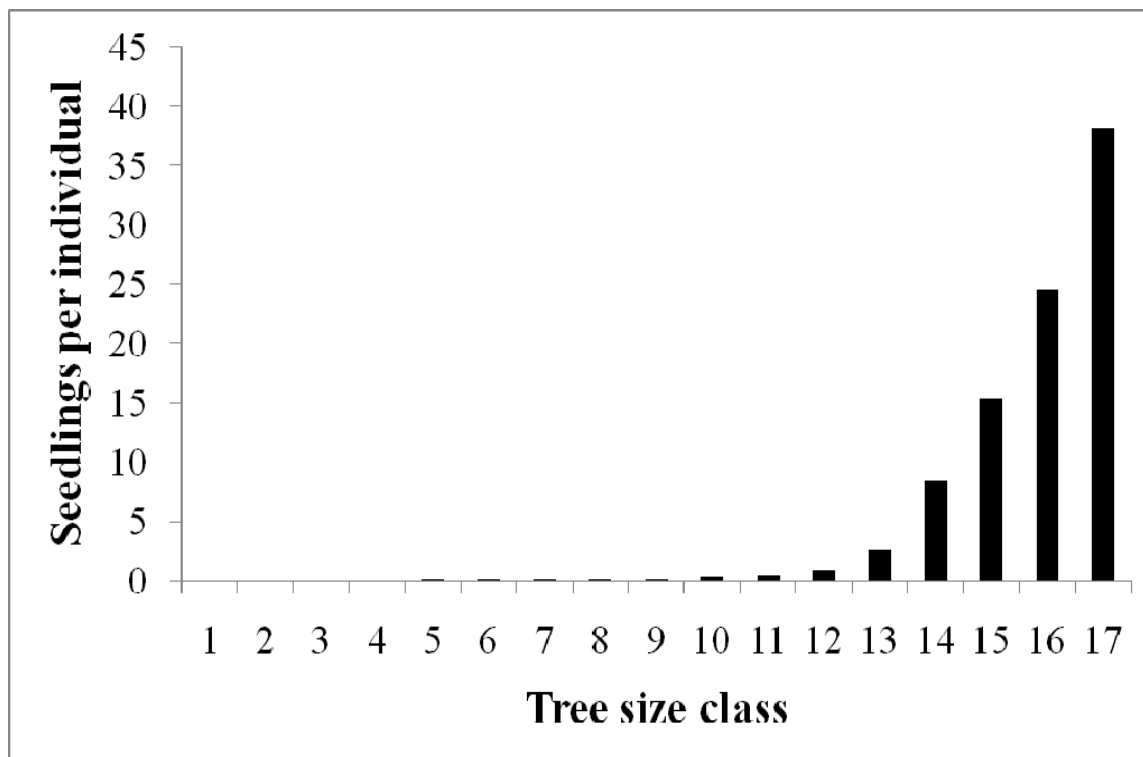


Figure 2-3: The average relative contribution of recruits per individual within each size class appearing in the following year.

Table 2-1: Parameter estimates for matrix model size classes: In all, 17 representative size classes were defined, with individuals ranging from 3 cm tall to > 300 cm tall and 98.1 cm diameter at breast height (DBH, 1.4 meters). 492 and 341 trees were used to estimate the life parameters of size classes 1-4 and 5 – 17, respectively. Columns 2 through 8 represent characteristics defining the decided upon size classes (St = stasis, G = growth, M = mortality, R = average contribution to recruits in next year per tree within size class). Column 9 represents the number of data points used to estimate the growth parameters of each size class; however it is important to note that due to the nature of the demographic data set an individual plant may be represented more than once in one or more size classes.

Size class	Height (cm)	DBH (cm)	St (%)	G (%)	M (%)	R (#)	Mean cones	N years of data
1	<15	N/A	0.111	0.061	0.828	0	0	198
2	15 - < 30	N/A	0.341	0.153	0.506	0	0	249
3	30 - < 47	N/A	0.383	0.188	0.430	0	0	128
4	47 – <140	N/A	0.736	0.107	0.157	0	0	121
5	≥ 140	0.4 – < 1.9	0.693	0.231	0.076	0.0032	0.01	515
6	≥ 140	1.9 - < 3.5	0.774	0.185	0.040	0.0217	0.07	372
7	≥ 140	3.5 – < 5.0	0.730	0.206	0.064	0.0407	0.14	141
8	≥ 140	5.0 - < 6.5	0.797	0.162	0.041	0.1050	0.35	74
9	≥ 140	6.5 - < 8.1	0.762	0.190	0.048	0.0636	0.21	36
10	≥ 140	8.1 - < 9.7	0.636	0.363	0.000	0.3302	1.1	23
11	≥ 140	9.7 - < 12.5	0.750	0.250	0.000	0.4166	1.4	46
12	≥ 140	12.5 - < 17	0.875	0.063	0.063	0.9253	3.1	57
13	≥ 140	17 - < 31.1	0.750	0.125	0.125	2.5916	8.7	42
14	≥ 140	31.1 - < 42	0.942	0.058	0.000	8.4888	28.5	68
15	≥ 140	42 - < 55.0	0.938	0.042	0.021	15.3560	51.6	54
16	≥ 140	55 - < 61.4	0.951	0.024	0.024	24.6004	82.7	48
17	≥ 140	61.4 <	0.955	0.000	0.045	38.1266	128.1	84

Table 2-2 – Finite growth rate, λ , for all 18 projected models including the upper and lower 95% confidence intervals (95% CI). The observed forest model estimate of λ was determined without any modifications to size class parameter estimates of mortality. Models A and B remove the common mycorrhizal network (CMN) benefit of 14.4% and 26.6% to increased survivorship from aged one and two individuals regardless of size or from size class one and two regardless of age, respectively. Each following model incrementally removes the Booth and Hoeksema (2010) estimated 2nd year survival reduction of 26.6% from a progressively higher size class, maintaining those modifications to survivorship made for model A and model B, until the entire tree life cycle has had the CMN removed.

Model	95% CI, lower bound of finite growth rate, λ	Finite growth rate, λ	95% CI, upper bound of finite growth rate, λ
Observed forest Data	0.9684	0.9822	0.9985
Non-CMN Ages 1 + 2 (Model A)	0.9662	0.9789	0.9963
Non-CMN Size class 1 + 2 (Model B)	0.9650	0.9778	0.9957
Non-CMN up to size class 3	0.9412	0.9750	0.9943
Non-CMN up to size class 4	0.9412	0.9707	0.9922
Non-CMN up to size class 5	0.9412	0.9679	0.9913
Non-CMN up to size class 6	0.9412	0.9657	0.9907
Non-CMN up to size class 7	0.9412	0.9648	0.9906
Non-CMN up to size class 8	0.9412	0.9643	0.9906
Non-CMN up to size class 9	0.9410	0.9641	0.9906
Non-CMN up to size class 10	0.9406	0.9641	0.9906
Non-CMN up to size class 11	0.9405	0.9640	0.9906
Non-CMN up to size class 12	0.9405	0.9640	0.9906
Non-CMN up to size class 13	0.9405	0.9640	0.9906
Non-CMN up to size class 14	0.9405	0.9640	0.9905
Non-CMN up to size class 15	0.9286	0.9640	0.9904
Non-CMN up to size class 16 (Model X)	0.9167	0.9640	0.9903
Non-CMN up to size class 17 (Model Y)	0.6944	0.7217	0.8333

Table 2-3 – Size class distribution of pines within the 2009 census compared to the estimated stable size class structure. * = rescaled estimates of pines <140 cm as determined by multiplying observed estimates by 10/3 to account for the adult skewed sampling scheme within the mixed size class plot (described in methods). DBH = diameter at breast height, 1.4 meters.

Tree size class	2009 census (#)	2009 census (%)	Estimated proportional stable size class structure
0.0 – < 15 cm tall	36.67*	0.058*	0.8183
15 – < 30 cm tall	40*	0.063*	0.0775
30 – < 47 cm tall	56.67*	0.089*	0.0204
47 – < 140 cm tall	283.3*	0.445*	0.0155
0.4 - < 1.9 cm DBH	44	0.069	0.0058
1.9 - < 3.5 cm DBH	76	0.119	0.0064
3.5 - < 5.0 cm DBH	43	0.068	0.0047
5.0 - < 6.5 cm DBH	21	0.033	0.0052
6.5 - < 8.1 cm DBH	8	0.013	0.0032
8.1 - < 9.7 cm DBH	2	0.003	0.0017
9.7 - < 12.5 cm DBH	2	0.003	0.0030
12.5 - < 17.0 cm DBH	2	0.003	0.0052
17.0 - < 31.1 cm DBH	2	0.003	0.0042
31.1 - < 42.0 cm DBH	5	0.008	0.0053
42.0 - < 55.0 cm DBH	7	0.011	0.0082
55.0 - < 61.4 cm DBH	5	0.008	0.0047
> 61.4 cm DBH	3	0.005	0.0107

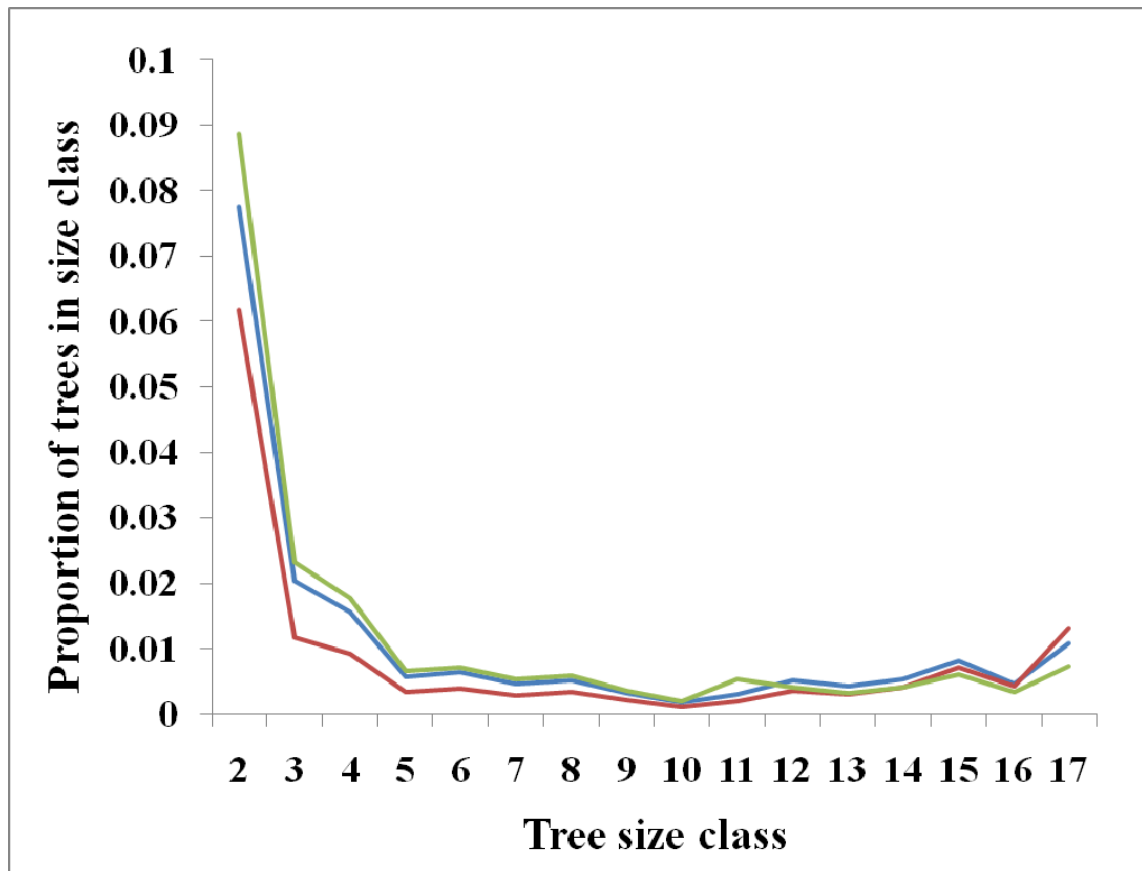


Figure 2-4: Proportional distribution of individuals within projected stable size class structure (Not depicted is size class 1, but see below for value). These three models were chosen as they represent the extremes observed for stable size class structures of all 18 models. Depicted are the observed forest data model (1st size class = .8183, blue line), the model removing the common mycorrhizal network benefit for all but the last size class (1st size class = .8986, model Y, red line), and the final model removing the common mycorrhizal network mortality reduction for all size classes (1st size class = .8067, model X, green line).

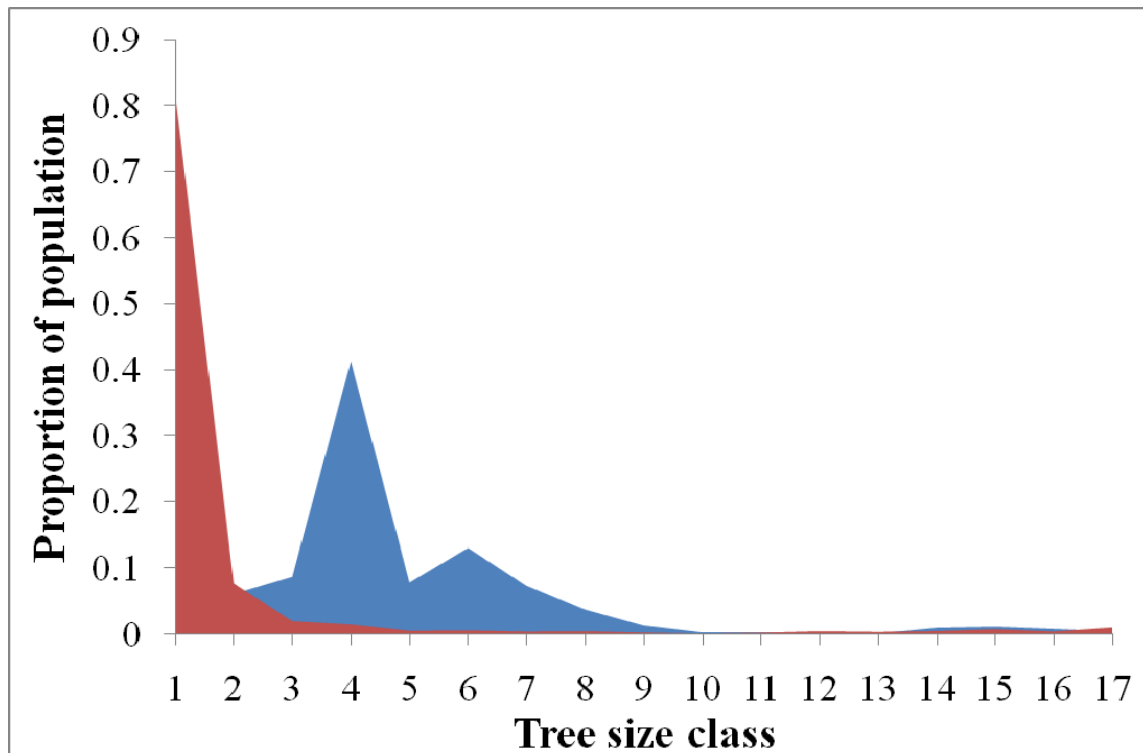


Figure 2-5 – Proportion of population within each size class for model estimated stable size class distributions for observed forest data (shaded red) versus the size class distribution for the last demographic census in 2009 (shaded blue).

Table 2-4: Elasticity analysis of observed demographic data reflecting the relative effect a small perturbation to the projection matrix affect the population growth rate. Elasticity values of fecundity (F), stasis (St), and growth (G) sum to a total of 1; however, in this table they may not due to rounding. Elasticity of survivorship is the sum of stasis and growth. Due to variation in the duration of time spent in a size class, values for survivorship may not be comparable between size classes. Therefore, unadjusted survivorship elasticities (St + G) have been divided by the maximum residence time (MRT) in each size class to give the adjusted elasticity for annual survivorship. * = Calculated by summing the estimated maximum number of years a seedlings is likely to remain in this size class

Size class	F	St	G	Unadjusted Survivorship	MRT	Adj. annual survivorship
1	0.0000	0.0014	0.0078	0.0091	4.0*	0.0023
2	0.0000	0.0045	0.0078	0.0123	5.0*	0.0025
3	0.0000	0.0050	0.0078	0.0128	1.62	0.0079
4	0.0000	0.0232	0.0078	0.0310	3.79	0.0082
5	0.0000	0.0186	0.0078	0.0264	3.26	0.0081
6	0.0000	0.0289	0.0078	0.0367	4.42	0.0083
7	0.0000	0.0225	0.0078	0.0303	3.70	0.0082
8	0.0000	0.0334	0.0078	0.0412	4.93	0.0084
9	0.0000	0.0215	0.0078	0.0293	3.59	0.0082
10	0.0000	0.0105	0.0078	0.0183	2.30	0.0080
11	0.0000	0.0412	0.0078	0.0490	3.75	0.0131
12	0.0001	0.0410	0.0077	0.0487	5.78	0.0084
13	0.0001	0.0426	0.0076	0.0502	5.99	0.0084
14	0.0005	0.1218	0.0070	0.1288	13.51	0.0095
15	0.0014	0.1681	0.0056	0.1737	17.86	0.0097
16	0.0013	0.0771	0.0043	0.0814	12.05	0.0068
17	0.0043	0.2129	0.0000	0.2129	27.77	0.0077

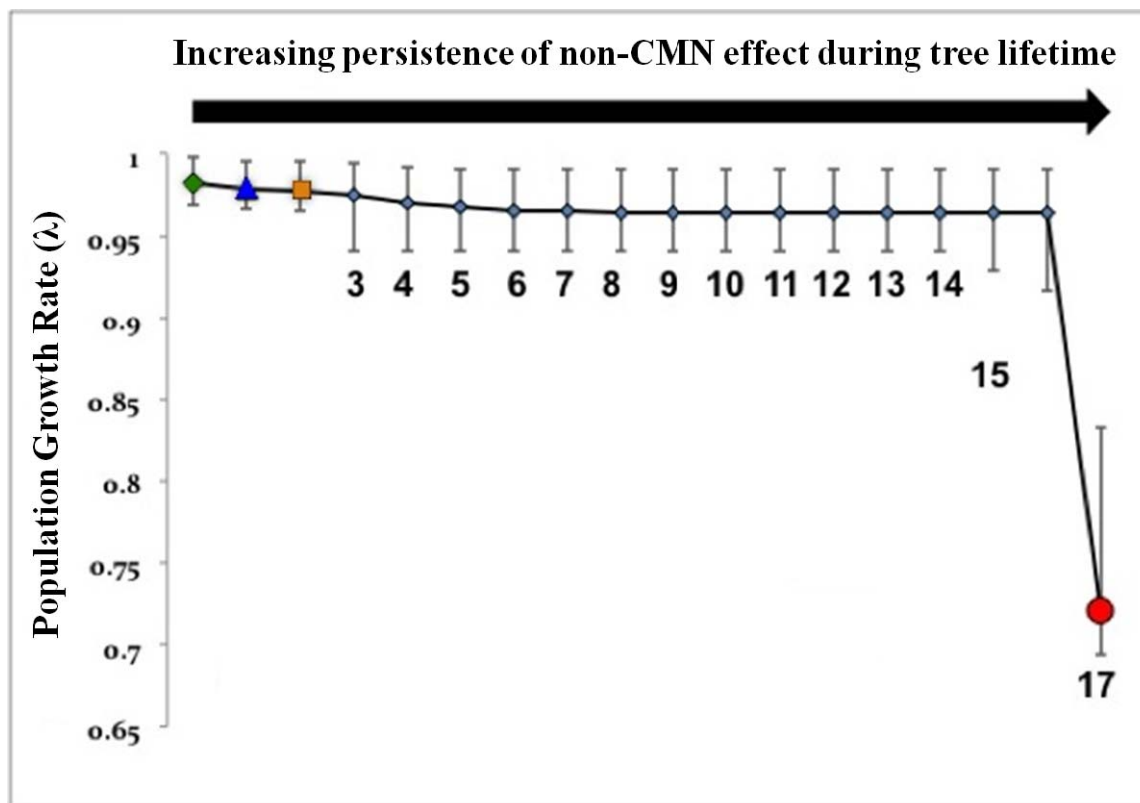


Figure 2-6: Projected population growth rates, λ , of the 18 projection matrices. From left to right include model estimates of λ for observed forest data ($\lambda=0.982$, 95% CI ± 0.998 , 0.968, Green diamond), removal of common mycorrhizal network (CMN) benefit to survivorship (14.4%, 26.6%) from seedlings age one and two ($\lambda=0.979$, 95% CI ± 0.996 , 0.966, Yellow triangle), removal of CMN benefit (14.4%, 26.6% of first and second size class ($\lambda=0.978$, 95% CI ± 0.995 , 0.965, Orange square). Each following point with associated number represents the model estimate of λ with CMN benefit to survivorship (26.6%) removed up to that numbered size class except for first size class which had only a 14.4% benefit to seedling survivorship removed. The final point estimate of λ ($\lambda=0.694$, 95% CI ± 0.833 , 0.721, Red circle) to the far right represents removal of the CMN benefit from the entire life cycle of the tree. Error bars are $\pm 95\%$ confidence intervals.

BIBLIOGRAPHY

- Acuna, E., M. Espinosa, J. Cancino, and R. Rubilar. 2010. Estimating the bioenergy potential of *Pinus radiata* plantations in Chile. *Ciencia E Investigacion Agraria* **37**:93-102.
- Agee, J. 1998. The landscape ecology of western forest fire regimes. *Northwest Science* **72**:24-34.
- Agerer, R. 2001. Exploration types of ectomycorrhizae: A proposal to classify ectomycorrhizal mycelial systems according to their patterns of differentiation and putative ecological importance. *Mycorrhiza* **11**:107-114.
- Allen, M. F. and M. G. Boosalis. 1983. Effects of 2 species of VA-mycorrhizal fungi on drought tolerance of winter-wheat. *New Phytologist* **93**:67-76.
- Aponte, C., L. Garcia, Maranon, and M. Gardes. 2010. Indirect host effect on ectomycorrhizal fungi: Leaf fall and litter quality explain changes in fungal communities on the roots of co-occurring Mediterranean oaks. *Soil Biology & Biochemistry* **42**:788-796.
- Ashkannejhad, S. and T. R. Horton. 2005. Ectomycorrhizal ecology under primary succession on coastal sand dunes: interactions involving *Pinus contorta*, suilloid fungi and deer. *New Phytologist* **169**:345-354.
- Baar, J., T. R. Horton, A. M. Kretzer, and T. D. Bruns. 1999. Mycorrhizal colonization of *Pinus muricata* from resistant propagules after a stand-replacing wildfire. *New Phytologist* **143**:409-418.
- Baker, A. and B. Murray. 2010. Relationships between leaf-litter traits and the emergence and early growth of invasive *Pinus radiata* seedlings. *Weed Research* **50**:586-596.
- Belling-Abler, R. A. 2004. Trace metal effects on ectomycorrhizal growth, diversity, and colonization of host seedlings. Virginia, Blacksburg.
- Bidartondo, M. I., J. Baar, and T. D. Bruns. 2001. Low ectomycorrhizal inoculum potential and diversity from soils in and near ancient forests of bristlecone pine (*Pinus longaeva*). *Canadian Journal of Botany* **97**:293-299.
- Blaudez, D., C. Jacob, K. Turnau, J. Colpaert, U. Ahonen-Jonnarth, R. D. Finlay, B. Botton, and M. Chalot. 2000. Differential responses of ectomycorrhizal fungi to heavy metals in vitro. *Mycological Research* **104**:1366-1371.
- Booth, M. G. 2004. Mycorrhizal networks mediate overstorey-understorey competition in a temperate forest. *Ecology Letters* **7**:538-546.
- Booth, M. G. and J. D. Hoeksema. 2010. Mycorrhizal networks counteract competitive effects of canopy trees on seedling survival. *Ecology* **91**:2294-2302.
- Borowicz, V. A. 2001. Do arbuscular mycorrhizal fungi alter plant-pathogen relations? *Ecology* **82**:3057-3068.
- Bowen, G. 1973. Mineral nutrition of mycorrhizas. Pages 151-201 in G. C. Marks, Kozolowski, TT., editor. *Ectomycorrhizas*. Academic Press, New York, USA.
- Boyd, R., R. Furbank, and D. Read. 1986. Ectomycorrhiza and the water relation of trees. Physiological and genetical aspects of mycorrhizae. Pages 689-693 in V. Gianinazzi-Pearson and V. Gianinazzi, editors. *Physiological and genetical aspects of mycorrhizae*, Paris, France.
- Bruns, T. D., J. Baar, P. Grogan, T. R. Horton, A. M. Kretzer, D. Redecker, J. Tan, and D. L. Taylor. 2005. Community dynamics of ectomycorrhizal fungi following the Mt. Vision fire. Pages 33-40 *Lessons learned from the October 1995 Mt. Vision Fire*. Points Reyes National Seashore, California.

- Canestro, D. Pers. Comm.
- Carpenter, E. J. and E. R. Storie. 1933. Soil survey of the San Luis Obispo area, California. United States Department of Agriculture, Bureau of Chemistry and Soils, Washington, D.C.
- Caswell, H. 2000. Matrix Population Models: Construction, Analysis, and Interpretation. 2nd edition. Sinauer, Sunderland, Massachusetts, USA.
- Corguz, V., M. Tabakovic-Tosic, M. Veselinovic, V. Raicevic, D. Drazic, L. Jovanovic, and D. Kikovic. 2010. The influence of heavy metals on the growth of ectomycorrhizal fungi. *Minerva Biotechnologica* **22**:17-22.
- de Kroon, H., A. Plaisier, J. van Groenendael, and H. Caswell. 1986. Elasticity: The relative contribution of demographic parameters to population growth rate. *Ecology* **67**:1427 - 1431.
- Deacon, J. W. and L. V. Fleming. 1992. Interactions of Ectomycorrhizal fungi. Pages 249-300 in M. F. Allen, editor. Mycorrhizal functioning. Chapman and Hall, New York, London.
- Doak, D., K. Gross, and W. F. Morris. 2005. Understanding and predicting the effects of sparse data on demographic analyses. *Ecology* **86**:1154-1163.
- Douglas, R. B., V. T. Parker, and K. W. Cullings. 2005. Belowground ectomycorrhizal community structure of mature lodgepole pine and mixed conifer stands in Yellowstone National Park. *Forest Ecology and Management* **208**:303-317.
- Dzierzon, H. and E. Mason. 2006. Towards a nationwide growth and yield model for radiata pine plantations in New Zealand. *Canadian Journal of Forest Research* **36**:2533-2543.
- Egerton-Warburton, L. M., J. I. Querejeta, and M. F. Allen. 2007. Common mycorrhizal networks provide a potential pathway for the transfer of hydraulically lifted water between plants (vol 58, pg 1473, 2007). *Journal of Experimental Botany* **58**:3484-3484.
- Fenton, G. R. 1951. Regeneration of *Pinus radiata*. *Forest Research Notes* **1**:1-14.
- Fleming, L., J. Deacon, and F. and Last. 1985. Ectomycorrhizal succession in a Scottish birchwood. Pages 259-264 in V. Gianinazzi-Pearson, editor. Physiological and Genetical Aspects of Mycorrhizae. INRA, Dijon, France.
- Fleming, L. V. 1983. Succession of mycorrhizal fungi on birch: infection of seedlings planted around mature trees. *Plant and Soil* **71**:263-267.
- Fomina, M., I. Alexander, J. Colpaert, and G. Gadd. 2005a. Solubilization of toxic metal minerals and metal tolerance of mycorrhizal fungi. *Soil Biology & Biochemistry* **37**:851-866.
- Fomina, M., E. Burford, and G. Gadd. 2005b. Toxic Metals and Fungal Communities. Pages 733-758 in J. F. W. Jon Dighton, and Peter Oudemans, editor. *The Fungal Community: Its Organization in the Ecosystem*. CRC Press.
- Fox, F. M. 1986. Groupings of ectomycorrhizal fungi of birch and pine, based on establishment of mycorrhizas on seedlings from spores in unsterile soils. *Transactions of the British Mycological Society* **87**:371-380.
- Frank, A. B. 1885. Über die auf Wurzelsymbiose beruhende Ernährung gewisser Bäume durch unterirdische Pilze. *Berichte der Deutschen Botanischen Gesellschaft* **3**:128-145.

- Frank, A. B. 2005. On the nutritional dependence of certain trees on root symbiosis with belowground fungi (an English translation of A.B. Frank's classic paper of 1885). *Mycorrhiza* **15**:267-275.
- Frazer, G. W., C. D. Canham, and K. P. Lertzman. 1999. Gap Light Analyzer (GLA), Version 2.0: Imaging software to Extract Canopy Structure and Gap Light Transmission Indices from True-color Fisheye Photographs, Users Manual and Program Documentation. Simon Fraser University/Institution of Ecosystem Studies, Burnaby, BC/ Millbrook/NY.
- Garbaye, J. 1994. Helper Bacteria: A new dimension to the mycorrhizal symbiosis. *New Phytologist* **128**:197-210.
- Garbaye, J. and R. Duponnois. 1992. Specificity and function of mycorrhization helper bacteria (MHB) associated with the *Pseudotsuga menziesii* - *Laccaria laccata* symbiosis. *Symbiosis* **14**:335-344.
- Gardes, M. and T. D. Bruns. 1993. ITS primers with enhanced specificity for basidiomycetes - application to the identification of mycorrhizae and rusts. *Molecular Ecology* **2**:113-118.
- Gardes, M. and T. D. Bruns. 1996. Community structure of ectomycorrhizal fungi in a *Pinus muricata* forest: above- and below-ground views. *Canadian Journal of Botany* **74**:1572 - 1583.
- Gherbi, H., C. Delaruelle, M. A. Selosse, and F. Martin. 1999. High genetic diversity in a population of the ectomycorrhizal basidiomycete *Laccaria amethystina* in a 150-year-old beech forest. *Molecular Ecology* **8**:2003 - 2013.
- Gomez, P., R. Bustamante, J. San Martin, and S. Hahn. 2011. Population structure of *Pinus radiata* D. Don in fragments of Maulino Forest in Central Chile. *Gayana Botanica* **68**.
- Gower, S. T., R. McMurtrie, and D. Murty. 1996. Aboveground net primary production decline with stand age: potential causes. *Trends in Ecology & Evolution* **11**:378-382.
- Grayston, S. J. and D. V. D. Jones. 1996. Rhizosphere carbon flow in trees, in comparison with annual plants: the importance of root exudation and its impact on microbial activity and nutrient availability. *Applied Soil Ecology* **5**:29--56.
- Grogan, P., J. Baar, and T. D. Bruns. 2000. Below-ground ectomycorrhizal community structure in a recently burned bishop pine forest. *Journal of Ecology* **88**:1051-1062.
- Hartnett, D. C. and G. W. T. Wilson. 1999. Mycorrhizae influence plant community structure and diversity in tallgrass prairie. *Ecology* **80**:1187-1195.
- Haywood, A. 2009. Estimation of height growth patterns and site index curves for *Pinus radiata* plantations in New South Wales, Australia. *Southern Forests* **71**:11-19.
- He, X.-H., C. Critchley, and C. Bledsoe. 2003. Nitrogen transfer within and between plants Through common mycorrhizal networks (CMNs). *Critical Reviews in Plant Sciences* **22**:531-567.
- He, X.-H., C. Critchley, H. Ng, and C. Bledsoe. 2005. Nodulated N₂-fixing *Casuarina cunninghamiana* is the sink for net N transfer from non-N₂-fixing *Eucalyptus maculata* via an ectomycorrhizal fungus *Pisolithus* sp. using ¹⁵NH₄⁺ or ¹⁵NO₃⁻ supplied as ammonium nitrate. *New Phytologist* **167**:897-912.
- Helm, D. J., E. B. Allen, and J. M. Trappe. 1996. Mycorrhizal chronosequence near Exit Glacier, Alaska. *Canadian Journal of Botany* **74**:1496-1506.

- Hoeksema, J. D., V. B. Chaudhary, C. A. Gehring, N. C. Johnson, J. Karst, R. T. Koide, A. Pringle, C. Zabinski, J. D. Bever, J. C. Moore, G. W. T. Wilson, J. N. Klironomos, and J. Umbanhowar. 2010. A meta-analysis of context-dependency in plant response to inoculation with mycorrhizal fungi. *Ecology Letters* **13**:394-407.
- Hood, G. 2010. PopTools. <http://www.poptools.org>.
- Horton, T., T. Bruns, and V. Parker. 1999. Ectomycorrhizal fungi associated with *Arctostaphylos* contribute to *Pseudotsuga menziesii* establishment. *Canadian Journal of Botany* **77**:93-102.
- Horton, T. R. 2002. Molecular approaches to ectomycorrhizal diversity studies: variation in ITS at a local scale. *Plant and Soil* **244**:29-39.
- Horton, T. R. and T. D. Bruns. 1998. Multiple-host fungi are the most frequent and abundant ectomycorrhizal types in a mixed stand of Douglas-fire (*Pseudotsuga menziesii*) and bishop pine (*Pinus muricata*). *New Phytologist* **139**:331-339.
- Horton, T. R. and T. D. Bruns. 2001. The molecular revolution in ectomycorrhizal ecology: peeking into the black-box. *Molecular Ecology* **10**:1855-1871.
- Horton, T. R., E. Cazares, and T. D. Bruns. 1998. Ectomycorrhizal, vesicular-arbuscular and dark septate fungal colonization of bishop pine (*Pinus muricata*) seedlings in the first 5 months of growth after wildfire. *Mycorrhiza* **8**:11-18.
- Huang, X. and A. Madan. 1999. Cap3: A DNA sequence assembly program. *Genome Research* **9**:868-877.
- Izzo, A., J. Agbowo, and T. D. Bruns. 2005. Detection of plot-level changes in ectomycorrhizal communities across years in an old-growth mixed-conifer forest. *New Phytologist* **166**:619-630.
- Johnson, E. A. and K. Miyanishi. 2008. Testing the assumptions of chronosequences in succession. *Ecology Letters* **11**:419-431.
- Johnson, N., J. Graham, and F. Smith. 1997. Functioning of mycorrhizal associations along the mutualism-parasitism continuum. *New Phytologist* **135**:575-586.
- Jumpponen, A. 2003. Soil fungal community assembly in a primary successional glacier forefront ecosystem as inferred from rDNA sequence analyses. *New Phytologist* **158**:569-578.
- Karst, J., L. Marczak, M. D. Jones, and R. Turkington. 2008. The mutualism-parasitism continuum in ectomycorrhizas: A quantitative assessment using meta-analysis. *Ecology* **89**:1032-1042.
- Keizer, P. J. and E. Arnolds. 1994. Succession of ectomycorrhizal fungi in roadside verges planted with common oak (*Quercus robur* L.) in Drenthe, The Netherlands. *Mycorrhiza* **4**:147-159.
- Kennedy, P. G., A. D. Izzo, and T. D. Bruns. 2003. There is high potential for the formation of common mycorrhizal networks between understorey and canopy trees in a mixed evergreen forest. *Journal of Ecology* **91**:1071 - 1080.
- Kennedy, P. G., K. G. Peay, and T. D. Bruns. 2009. Root tip competition among ectomycorrhizal fungi: Are priority effects a rule or an exception? *Ecology* **90**:2098-2107.
- Kernaghan, G., P. Widden, Y. Bergeron, S. Legare, and D. Pare. 2003. Biotic and abiotic factors affecting ectomycorrhizal diversity in boreal mixed-woods. *Oikos* **102**:497-504.

- Kjoller, R. and T. D. Bruns. 2003. Rhizopogon spore bank communities within and among California pine forests. *Mycologia* **95**:603-613.
- Klironomos, J., J. McCune, M. Hart, and J. Neville. 2000. The influence of arbuscular mycorrhizae on the relationship between plant diversity and productivity. *Ecology Letters* **3**:137-141.
- Klironomos, J., M. Zobel, M. Tibbett, W. D. Stock, M. C. Rillig, J. L. Parrent, M. Moora, A. Koch, J. Facelli, E. Facelli, I. A. Dickie, and J. D. Bever. 2011. Forces that structure plant communities: quantifying the importance of the mycorrhizal symbiosis. *New Phytologist* **189**:366-370.
- Koide, R. T., P. E. Courty, and J. Garbaye. 2007. Research perspectives on functional diversity in ectomycorrhizal fungi. *New Phytologist* **174**:240-243.
- Kranabetter, J. M., J. Friesen, S. Gamiet, and P. Kroeger. 2005. Ectomycorrhizal mushroom distribution by stand age in western hemlock - lodgepole pine forests of northwestern British Columbia. *Canadian Journal of Forest Research* **35**:1527-1539.
- Last, F., J. Dighton, and P. and Mason. 1987. Successions of sheathing mycorrhizal fungi. *Trends in Ecology and Evolution* **2**:157-161.
- Lilleskov, E. A. and T. D. Bruns. 2003. Root colonization dynamics of two ectomycorrhizal fungi of contrasting life history strategies are mediated by addition of organic nutrient patches. *New Phytologist* **159**:141-151.
- Marcos, J. A., E. Marcos, A. Taboada, and R. Tarrega. 2007. Comparison of community structure and soil characteristics in different aged *Pinus sylvestris* plantations and a natural pine forest. *Forest Ecology and Management* **247**:35-42.
- Martin, K. J. and P. T. Rygielwicz. 2005. Fungal-specific PCR primers developed for analysis of the ITS region of environmental DNA extracts. *BMC Microbiology* **5**:28-39.
- Mason, P. A., J. Wilson, F. T. Last, and C. Walker. 1983. The concept of succession in relation to the spread of sheathing mycorrhizal fungi on inoculated tree seedlings growing in unsterile soils *Plant and Soil* **71**:247-256.
- McDonald, P. M. and R. J. Laacke. 1990. *Pinus radiata* D. Don Monterey Pine. Pages 433-441 in R. M. Burns and B. H. Honkala, editors. *Silvics of North America. Volume 1. Conifers*. United States Department of Agriculture, Forest Service, Washington, D.C.
- Moloney, K. 1986. A Generalized Algorithm for Determining Category Size. *Oecologia* **69**:176-180.
- Mosquera-Losada, M., N. Ferreiro-Dominguez, and A. Rigueiro-Rodriguez. 2010. Fertilization in pastoral and *Pinus radiata* D. Don silvopastoral systems developed in forest and agronomic soils of Northwest Spain. *Agriculture Ecosystems & Environment* **139**:618-628.
- Nara, K., H. Nakaya, B. Wu, Z. Zhou, and T. Hogetsu. 2003. Underground Primary Succession of Ectomycorrhizal Fungi in a Volcanic Desert on Mount Fuji. *New Phytologist* **159**:743-756.
- Newton, A. 1992. Towards a functional classification of ectomycorrhizal fungi. *Mycorrhiza* **2**:75-79.
- O'brien, H. E., J. L. Parrent, J. A. Jackson, J.-M. Moncalvo, and R. Vilgalys. 2005. Fungal Community Analysis by Large-Scale Sequencing of Environmental Samples. *Applied and Environmental Microbiology* **71**:5544-5550.

- O'brien, M., K. O'Hara, N. Erbilgin, and D. Wood. 2007. Overstory and shrub effects on natural regeneration processes in native *Pinus radiata* stands. *Forest Ecology and Management* **240**:178-185.
- O'Connor, P., S. Simith, and E. Smith. 2002. Arbuscular mycorrhizas influence plant diversity and community structure in a semiarid herbland. *New Phytologist* **154**:209-218.
- Parke, J. L., R. G. Linderman, and C. H. Black. 1983. The role of ectomycorrhizas in drought tolerance of douglas-fir seedlings. *New Phytologist* **95**:83-95.
- Peay, K. G., P. G. Kennedy, and T. D. Bruns. 2010. Rethinking ectomycorrhizal succession: are root density and hyphal exploration types drivers of spatial and temporal zonation? *Fungal Ecology* **4**:233-240.
- Peng, C. 2000. Growth and yield models for uneven-aged stands: past, present and future. *Forest Ecology and Management* **132**:259-279.
- Perry, D. A., M. P. Amaranthus, J. G. Borchers, S. L. Borchers, and R. E. Brainerd. 1989. Bootstrapping in ecosystems. *Bioscience* **39**:230-237.
- Piirto, D. and S. Valkonen. 2005. Structure and stand development of pitch canker infected Monterey pine stands at Ano Nuevo, California. *Forest Ecology and Management* **213**:160-174.
- Read, D. J. 1991. Mycorrhizas in ecosystems. *Experientia* **47**:376-391.
- Read, D. J. and J. Perez-Moreno. 2003. Mycorrhizas and nutrient cycling in ecosystems - a journey towards relevance? *New Phytologist* **157**:475-492.
- Rogers, D. L. 2002. Selected biological features of Monterey pine and its forests. In situ genetic conservation of Monterey pine (*Pinus radiata* D. Don): information and recommendations. Report No. 26, Davis, CA.
- Rogers, D. L. 2004. In situ genetic conservation of a naturally restricted and commercially widespread species, *Pinus radiata*. *Forest Ecology and Management* **197**:311-322.
- Roy, D. 1966. Silvical characteristics of Monterey pine (*Pinus radiata* D. Don). in P. S. F. a. R. E. Station, editor. USDA Forest Service, Berkeley, CA.
- Rusca, T. A., T. A. Kennedy, and T. D. Bruns. 2006. The effect of different pine hosts on the sampling of *Rhizopogon* spore banks in five Eastern Sierra Nevada forests. *New Phytologist* **170**:551-560.
- Sikes, B. A. and J. N. Klironomos. 2009. Plant and fungal identity determines pathogen protection of plant roots by arbuscular mycorrhizas. *Journal of Ecology* **97**:1274-1280.
- Silvertown, J. and D. Charlesworth. 2001. *Introduction to Plant Population Biology*. 4th edition. Blackwell Science, Berlin, Germany.
- Silvertown, J., M. Franco, I. Pisanty, and A. Mendoz. 1993. Comparative plant demography - relative importance of life-cycle components to the finite rate of increase in woody and herbaceous perennials. *Journal of Ecology* **81**:465-476.
- Simard, S. W., D. Durall, and M. and Jones. 2002. Carbon and nutrient fluxes within and between mycorrhizal plants. Pages 33-74 in M. G. A. Van der Heijden and I. R. & Sanders, editors. *Mycorrhizal Ecology*. Springer-Verlag, New York.
- Simard, S. W. and D. M. Durall. 2004. Mycorrhizal networks: a review of their extent, function, and importance. *Canadian Journal of Botany* **82**:1140-1165.

- Simard, S. W., D. A. Perry, M. D. Jones, D. D. Myrold, D. M. Durall, and R. Molina. 1997. Net transfer of carbon between ectomycorrhizal tree species in the field. *Nature* **388**:579-582.
- Smith, J. E., R. Molina, M. M. P. Huso, D. L. Luoma, D. McKay, M. A. Castellano, T. Lebel, and Y. Valachovic. 2002. Species richness, abundance, and composition of hypogeous and epigeous ectomycorrhizal fungal sporocarps in young, rotation-age, and old-growth stands of Douglas-fir (*Pseudotsuga menziesii*) in the cascade range of Oregon, USA. *Canadian Journal of Botany* **80**:186-204.
- Smith, M., G. Douhan, and D. Rizzo. 2007. Ectomycorrhizal community structure in a xeric *Quercus* woodland based on rDNA sequence analysis of sporocarps and pooled roots. *New Phytologist* **174**:847-863.
- Smith, S. E. and D. J. Read. 2008. *Mycorrhizal symbiosis*. 3rd edition. Academic Press, Amsterdam, Netherlands.
- Stendell, E., T. Horton, and T. Bruns. 1999. Early effects of prescribed fire on the structure of the ectomycorrhizal fungus community in a Sierra Nevada ponderosa pine forest. *Mycological Research* **103**:1353-1359.
- Stephens, S., D. Piirto, and D. Caramagno. 2004. Fire regimes and resultant forest structure in the native Ano Nuevo Monterey Pine (*Pinus radiata*) Forest, California. *American Midland Naturalist* **152**:25-36.
- Sthultz, C., T. Whithas, K. Kennedy, R. Deckert, and C. Gehring. 2009. Genetically based susceptibility to herbivory influences the ectomycorrhizal fungal communities of a foundation tree species. *New Phytologist* **184**:657-667.
- Sylvia, D. M. 1983a. Role of *Laccaria laccata* in protecting primary roots of douglas-fir from root-rot. *Plant and Soil* **71**:299-302.
- Sylvia, D. M., Sinclair, W.A. 1983b. Suppressive influence of *Laccaria laccata* on *Fusarium oxysporum* and on Douglas fir seedlings. *Phytopathology* **73**:384-389.
- Taylor, D. L. and T. D. Bruns. 1999. Community structure of ectomycorrhizal fungi in a *Pinus muricata* forest: minimal overlap between the mature forest and resistant propagule communities. *Molecular Ecology* **8**:1837-1850.
- Taylor, D. L., I. C. Herriott, J. Long, and K. O'Neill. 2007. TOPO TA is A-OK: a test of phylogenetic bias in fungal environmental clone library construction. *Environmental Microbiology* **9**:1329-1334.
- Tedersoo, L., U. Koljalg, N. Hallenberg, and K. H. Larsson. 2003. Fine scale distribution of ectomycorrhizal fungi and roots across substrate layers including coarse woody debris in a mixed forest. *New Phytologist* **159**:153-165.
- Treseder, K. K. 2005. Unearthing ectomycorrhizal dynamics. *New Phytologist* **166**:358-359.
- Twieg, B. D. 2006. Ectomycorrhizal communities of Douglas-fir and paper birch along a gradient of stand age after clearcutting and wildfire in the Interior Cedar-Hemlock Zone, southern British Columbia. University of British Columbia, Vancouver.
- Twieg, B. D., D. M. Durall, and S. W. Simard. 2007. Ectomycorrhizal fungal succession in mixed temperate forests. *New Phytologist* **176**:437-447.
- van der Heijden, M. G. A., J. N. Klironomos, M. Ursic, P. Moutoglis, R. Streitwolf-Engel, T. Boller, A. Wiemken, and I. R. Sanders. 1998. Mycorrhizal fungal diversity determines plant biodiversity, ecosystem variability and productivity. *Nature* **396**:69-72.

- van der Heijden, M. G. A., R. Streitwolf-Engel, R. Riedl, S. Siegrist, A. Neudecker, K. Ineichen, T. Boller, A. Wiemken, and I. R. Sanders. 2006. The mycorrhizal contribution to plant productivity, plant nutrition and soil structure in experimental grassland. *New Phytologist* **172**:739-752.
- Vandermeer, J. 1978. Choosing Category Size in a Stage Projection Matrix. *Oecologia* **32**:79-84.
- Visser, S. 1995. Ectomycorrhizal Fungal Succession in Jack Pine Stands Following Wildfire. *New Phytologist* **129**:389-401.
- Walbert, K., T. D. Ramsfield, H. J. Ridgway, and E. E. Jones. 2010. Ectomycorrhiza of *Pinus radiata* (D. Don 1836) in New Zealand - an above- and below-ground assessment. *Australasian Mycologist* **29**:7-16.
- Wang, B. and Y. L. Qiu. 2006. Phylogenetic distribution and evolution of mycorrhizas in land plants. *Mycorrhiza* **16**:299-363.
- White, K. 1999. Revisiting native *Pinus radiata* forests after twenty-nine years. *Madrono* **46**:80-87.
- Williams, M. and G. Wardle. 2007. *Pinus radiata* invasion in Australia: Identifying key knowledge gaps and research directions. *Austral Ecology* **32**:721-739.
- Yamashita, S., K. Fukuda, and S. Ugawa. 2008. Ectomycorrhizal communities on tree roots and in soil propagule banks along a secondary successional vegetational gradient. *Forest Science* **53**:635-644.

VITA

Born in 1983, the second son of Ron and Joyce Hennig, Kristopher Jordan Hennig was raised in Eau Claire, Wisconsin. Through family outings and road trips in a beat up 1986 Ford F-150, later dubbed the “Grey Ghost”, he gained an appreciation for the natural world that manifested itself in his involvement in his high school ecology club at North High School and later by gaining a Biology degree with a minor in environmental sciences from the University of Wisconsin, Eau Claire. After a particularly devastating experience working at a fish processing plant in Oregon, he decided that it was time to investigate further education opportunities. Having worked on a prairie restoration project under the guidance of Dr. Evan Weiher, gaining experience working with arbuscular mycorrhizal fungi, and soon after reading various mycological texts, Kris sought to invest his time studying the fungal world, particularly that of the ectomycorrhizal fungi.